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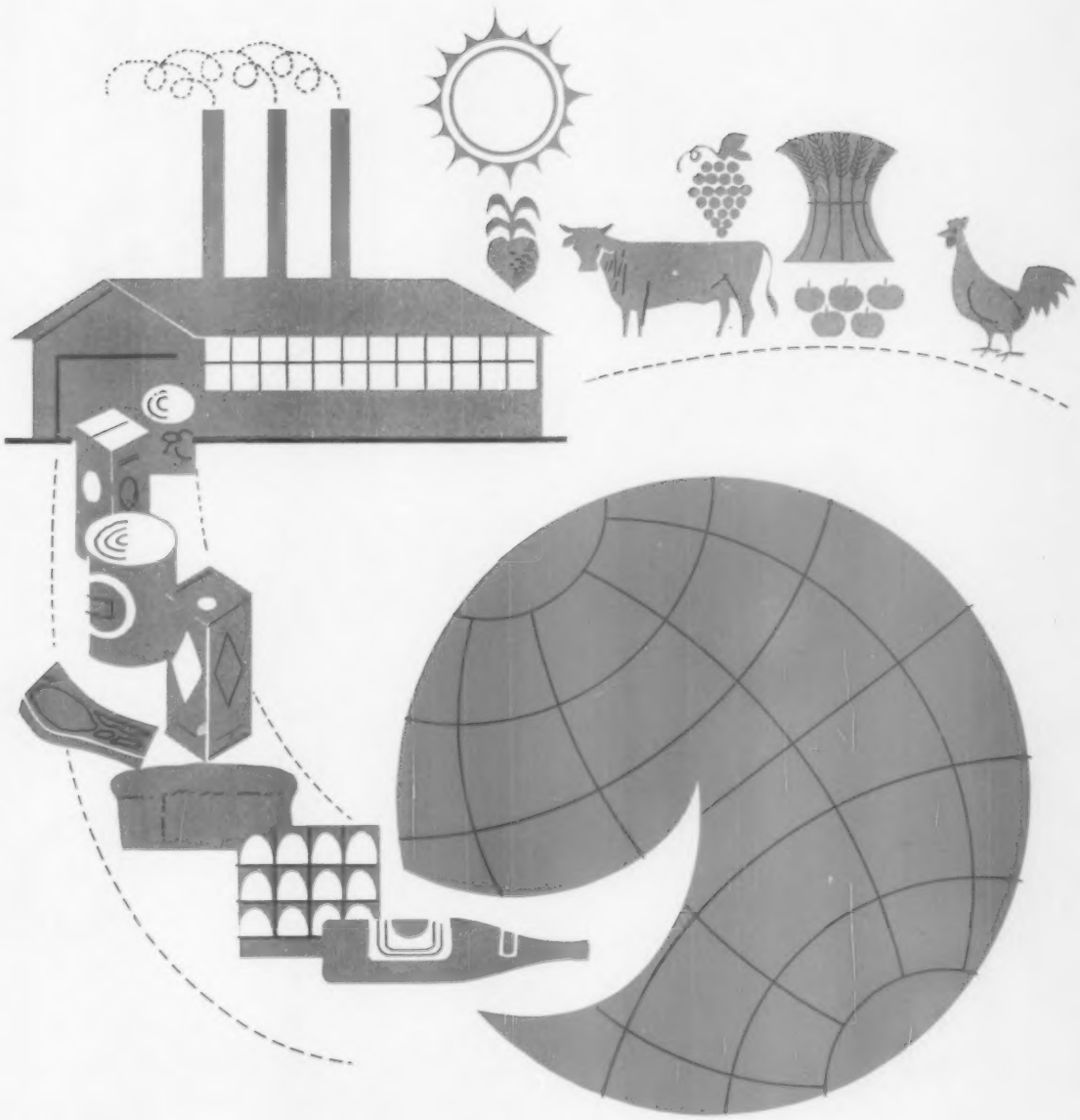

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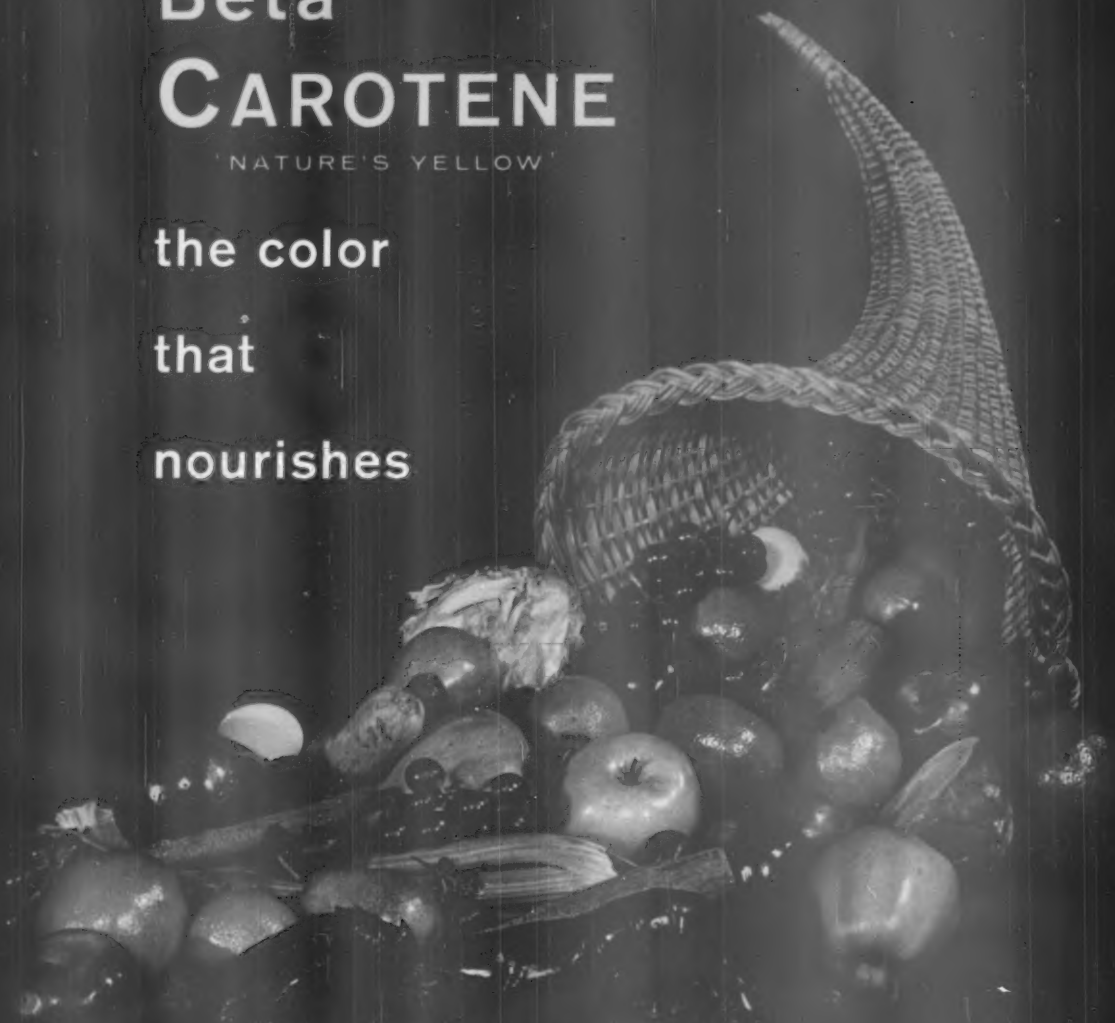

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Food Uses and Properties of Soybean Protein. I.

Food Uses[a]

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(Manuscript received June 28, 1960)

SUMMARY

Whole soybean may be baked, boiled, used for making sprouts, fresh and dried tofu, vegetable milk, yuba, and many fermented products. In the U. S. the trend had been to use a special grade of defatted soybean meal and of protein concentrates derived from the meal. These fractions include the acid-precipitated protein, soluble soybean proteinate, protein concentrate 70, aerating agents, soy flour, and alcohol-washed or Gelsoy-type products. These products are used not alone but combined in standard-type food products for their functional effect or for improvement of protein nutrition. Food use of U. S. soybeans is expanding more rapidly in foreign countries than domestically.

Soybean oil meal (SOM) is produced in the U. S. at the rate of 9.5 million tons annually. It is used mainly for animal feeds, but specially prepared grades of the meal are the raw materials for a number of foods and industrial products, and for isolated soybean protein.

Commercial solvent-defatted SOM contains 44 or 50% (N × 6.25) and is sometimes called soybean protein. The higher protein level is obtained by removing the seed coat. The term "protein" may have originated in the industry because the value of the meal is based almost entirely on its protein content. Herein we limit "protein" to more or less purified proteins extracted from the dehulled and defatted meal.

An extensive review of soybeans as well as other sources of vegetable proteins for food use has appeared (Altschul, 1958); our review emphasizes the soybean proteins and protein concentrates used in foods. A review (Bain *et al.*, in press) emphasizing industrial rather than food use is to appear shortly.

COMMERCIAL SOYBEAN PROTEIN FRACTIONS

The commercial isolation of soybean protein has a number of variations; the two main steps, however, are extraction of dehulled and undenatured soybean flakes (Belter and Smith, 1952) with water or dilute alkali solution (Smith *et al.*, 1938, 1952; Smith and Circle, 1938) and precipitation of about 88% of the extracted nitrogen at pH 4.5 (Smith and Circle, 1939). Laboratory yield of acid-precipitated protein may be as much as 79% of the nitrogen in the defatted meal. A pilot plant for isolating soybean protein has been described by Belter *et al.* (1944).

The centrifugate from the acid-precipitated protein is called soybean whey solution. The nitrogen in the whey, although variable, is about half protein and half nonprotein. Thus whey protein is about 6% of the extracted nitrogen. Because of its low concentration, whey protein is not yet economically recoverable and is a waste disposal problem.

The third fraction, or "residue," in protein isolation accounts for 25–30% of the meal, and contains 25–32% protein and water-insoluble carbohydrates; little is known about the proteins or carbohydrates of this fraction.

Acid-precipitated protein produced for food and that for industrial purposes are markedly different. Food protein is processed with a minimum of chemical modification, whereas the industrial protein is modified by caustic and other chemicals that make it unsuitable for food use or nutritional studies.

SOYBEAN FOODS

Soybean varieties. The original breeding stock for U. S. soybeans came from the Orient (Carter and Hopper, 1942). This stock was divided into garden and field varieties. No definition is used to distinguish them, but there are marked differences. The garden varieties have a less-beany flavor and better texture, and are generally more acceptable for table use than field types (Woodruff and Klaas, 1938), which contain higher oil, yield better, harvest better, and have other properties important to commercial production. Because garden types are grown in small volume, any development of soybeans for food or industrial use will depend primarily on field varieties, of which we have 35 or more on the recommended list (Smith, 1959).

Field varieties differ considerably in color of seed coat, color of cotyledons, protein and oil content, and, to some degree, flavor, texture, and size. Differences in these properties and others are shown in research that compares U. S. and Japanese soybeans for making tofu (Smith *et al.*, 1960). Investigations relating variety to food uses are so few that about all we can say now is that we are aware of these differences, believe they are important, and feel further research is needed to select the most suitable varieties.

For a number of years we have had a very successful plant breeding program in this country, and each year several new improved varieties are released to replace an approximately equal number of less desirable older ones. Our breeding program has been concerned primarily with increasing soybean yields and raising the oil content; with soybean foods gaining more interest, however, we anticipate that plant breeders will give greater recognition to food properties in selecting new varieties.

[a] Presented at the Twentieth Annual Meeting of the Institute of Food Technologists, San Francisco, California, May 16–19, 1960.

[b] A laboratory of the Northern Utilization Research and Development Division, Agricultural Research Service, U. S. Department of Agriculture.

Trends in Protein Requirements

When soybeans were grown only in the Orient, they were used almost entirely as food. After they were introduced into the United States, many people assumed they would soon find a place in the American diet. This assumption was true for the oil, for about 90% of the processed oil now goes into food uses; in fact, soybean oil is the most important of our edible fats. Food markets for soybean meal products and isolated protein developed more slowly; these do not fit into our eating habits as easily as does the oil.

Increasing markets for SOM as an animal feed, as well as expanding export of soybeans from the United States, have had an important effect on the relative value of the oil and protein. Early in the history of soybean processing in the U. S. the oil had far more value than the meal or protein, which was often called a byproduct. Today, dollar value of the meal (48 lb) from a bushel of beans exceeds that of the oil (11 lb). Since the 1930's soybeans have become a major crop, oil prices have decreased from 20 cents or more per pound to less than 8 cents, and protein prices (in 44% protein meal), although variable, have increased from less than 2 cents to more than 6 cents per pound. Soybeans have never had a high support price; these changes result more from supply and demand than from regulations. The increased market for SOM was stimulated by nutritional research showing the economic advantages of high protein in animal feeds; this market, from the standpoint of balanced rations, is still under-supplied (McKinney and Cowan, 1956). Even though animal feeds are the most important outlet for domestic soybean protein, world food markets are becoming more important and are expected to expand.

About 143 million bushels of soybeans or approximately 30% of the beans moved off the farm are exported as well as an additional 30% of the oil (Hedge, 1960). Our largest customer is Japan, with a 1959 import estimated at more than 40 million bushels. These imports and her own production, about 15–18 million bushels, go into food where the protein is more important than the oil. Most U. S. beans imported into Japan are processed for oil and meal, and then the meal is used in high-protein foods. If we include foreign and domestic utilization, about 10% of the protein of the U. S. processed crop goes into food products—a significant factor in marketing soybeans.

World surveys on food requirements report adequate supplies of oil but a great shortage of protein (Foreign Agricultural Organization, 1948–1956); these reports suggest that increasing demand for protein in other countries as well as our own will likely cause a trend toward a further increase in the price of soybean protein over that of the oil. Since the U. S. grows about 55% (550 million bushels) of the world's supply of soybeans, and since some authorities predict that production will reach nearly a billion bushels in the next 10–15 years, we have a continuing interest in foreign markets.

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seeds. Their advantages are in the high nutritional value of their protein, their relatively high ratio of protein to oil, their low proportion of seed coat, and their low cost of production (resulting from our highly mechanized farming methods). These factors should make the market for soybean protein expand considerably.

Soybeans and Fractions Used in Food

Whole soybeans. The whole soybean may be baked or boiled, or used in making sprouts, fresh and dried tofu, vegetable milk, yuba, and many fermented products. Baked or boiled soybeans are not very popular in this country or in the Orient, probably because soaking and cooking require long periods, and the product causes flatulence. The sprouts, used as a green salad or fried, are a rich and nutritious food but are not produced commercially in this country; mung bean sprouts are more popular. Soybean milk (Fomon, 1959), produced on a very small scale in the U. S., is recommended for feeding babies that are allergic to cow's milk. The milk, prepared from a water extract of the ground whole bean, has a serious flavor problem and sometimes appears to cause digestive disturbance in weaning babies. These problems have stimulated investigation of new methods of preparing soya milk, using an oil emulsified in the isolated protein and containing added vitamins and minerals. Cooked whole beans are used very extensively in the Orient for fermentation into miso or soy paste, natto, hamannatto, shoyu (soy sauce), tempeh, and some less important foods (Smith, 1958). The modern trend in Japan in making shoyu is to replace whole beans with defatted soybean meal.

Defatted soybean meal. In this country the starting material for most soybean foods is dehulled, defatted, and substantially undenatured soybean meal containing 50% protein or more ($N \times 6.25$). Occasionally, soybeans are found that will produce a meal containing up to 60% protein. Made from specially prepared meal (Belter and Smith, 1952) are food-grade soybean protein isolates and concentrates such as a) soy flour, b) acid-precipitated protein, c) soluble soybean proteinate, d) 70% protein concentrate, e) aerating agents, f) Gelsoy-type products (Glabe *et al.*, 1956), and g) dry soybean whey solids. Each is made in several modifications, depending on the procedure selected by the processor and its intended use.

The protein materials described, however, are not used as foods alone but are added to other foods for functional effects or as a nutritional supplement. An example of a functional effect is that of soy flour in doughnuts (to control their absorption of fat). The protein solubility of soy flour, as controlled by heat treatment, is inversely related to fat absorption by the doughnut (Northern Regional and Central Research Laboratories, 1951). Another example is soy whip (Burnett and Gunther, 1949), used as an aerating agent in nougats and desserts. Other functional uses of soy flour, isolated protein, and other protein fractions are for bleaching wheat flour (Haas and Bohn, 1934); as thickening, emulsifying, and

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gelling agents, and meat binder (Frank and Circle, 1959; Johnson and Circle, 1959); and for binding water in foods (Glabe *et al.*, 1956).

The most important use of soybean proteins in foods is to improve nutritional value, particularly that of bread, breakfast foods, and other cereal products. Soybean protein, with its high-lysine content (6.5%), supplements the very low lysine in most cereals. When as little as 5% soy flour is added to patent wheat flour, the nutritive value of wheat protein is increased remarkably (Jones and Divine, 1944).

In 1935 (Bailey *et al.*, 1935) soy flour was recommended as an additive to bread, but its first use often resulted in disappointment because of poor color, flavor, and loss in loaf volume. The early processing of soy flour, made without any standardized controls, varied in composition and physical properties. Furthermore, there was no dependable information on how to use it. Since then, Ofelt *et al.* (1954), Finney (1945), and Pollock and Geddes (1960) have shown that raw (untoasted) soy flour causes softening of bread dough and loss of loaf volume. These effects may be caused by the amylases (Ofelt *et al.*, 1955a), proteases (Ofelt *et al.*, 1955b), or some other unidentified enzyme or reducing substance in the soy flour. Commercial soy flours, however, are always heated enough to inactivate all their enzymes substantially. Reduction in loaf volume is avoided by proper use of oxidizing agents (Ofelt *et al.*, 1954). When soy flour is properly used at a 5% level of the wheat flour, its presence in bread is not detectable by taste (Ofelt *et al.*, 1952).

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Isolated proteins. Acid-precipitated protein and water-soluble sodium proteinate have been available for industrial uses since about 1936, but food grades have been available for only a few years and have not yet developed a definite use pattern. Johnson and Circle (1959) enumerate about 90 foods that they believe these proteins can be used in, including bakery products, breakfast cereals, spaghetti, noodles, egg and dairy-type products, sausages and ground meats, edible coatings, Oriental-type foods, and specialties such as baby foods, candies, flavor carriers, gravies, emulsions, and geriatric food. Many of these uses are said to be under active investigation by food processors. There are many reasons for believing that isolated proteins will eventually find a wider and perhaps more extensive use than soy flour. Even though isolated protein may cost 3–5 times an equal amount of protein in the form of soy flour, it has advantages in certain applications.

In making isolated protein, almost all flavor is removed; part of it goes into the whey solution and part is destroyed or steam-distilled during drying. Also, the low-molecular-weight compounds that may interfere in some food preparations are removed in the whey, e.g., soybean components responsible for causing soft doughs (Ofelt *et al.*, 1954) and loss of loaf volume. In fact, acid-precipitated protein has very little known chemical reactivity in most foods. If desired, solubility can be obtained by using the sodium proteinate although it will be more reactive, especially to acids, metal ions, and other positively charged molecules.

Protein concentrate. Extraction of dehulled and defatted meal with dilute acid (pH 4.5) removes soluble sugars, nonprotein nitrogen, and other low-molecular-weight components and a small amount of protein. The flavors are also mostly removed in the extract or in drying. The dried concentrate contains about 70% protein unless soybeans containing above-average protein are used.

This product, having a manufacturing cost between that of soy flour and isolated protein, has been introduced recently into the food industry. This protein concentrate is a combination of the acid-precipitated protein plus the residue normally obtained in isolating the acid-precipitated protein. It resembles acid-precipitated protein more than soy flour and is expected to share some of the markets outlined for the acid-precipitated protein.

A protein concentrate can be made also by extraction of SOM with about 70% ethanol at 50°C or higher. This type of product is finding its place in the food industry.

Gelsoy, a bland-tasting water-soluble product (Glabe *et al.*, 1956) containing about 55% protein, is made by washing undenatured hexane-extracted flakes with ethanol, removing the ethanol by a flash process (Brekke *et al.*, 1959) extracting the alcohol-washed flakes with water, and recovering the ex-

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tracted material by spray drying. Gelsoy has emulsifying properties useful in canned and sausage-type meats, for controlling over-run in low-fat frozen confections, and as an aerating agent. Although not yet in commercial production in the U. S., its production was recently initiated in Japan, where extensive utilization is anticipated. The function of the 90–95% ethanol is to remove the bitter and beany flavor of hexane-extracted flakes. The flavor can be removed with methanol but somewhat more denaturation occurs than with the same concentration of ethanol. Use of 99% isopropanol improves the color of the flakes but not the flavor; however, on dilution of isopropanol to 85–90% it will also remove flavor. The most difficult step in making Gelosy is to remove the flavor without excessive loss in protein solubility.

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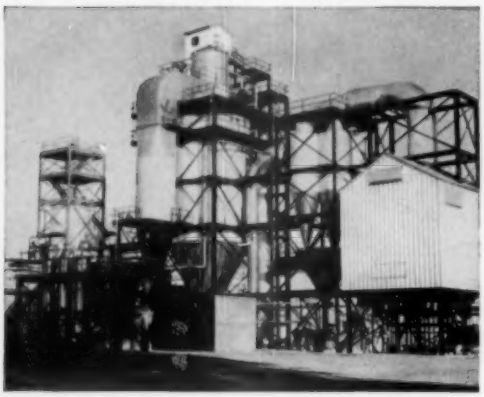

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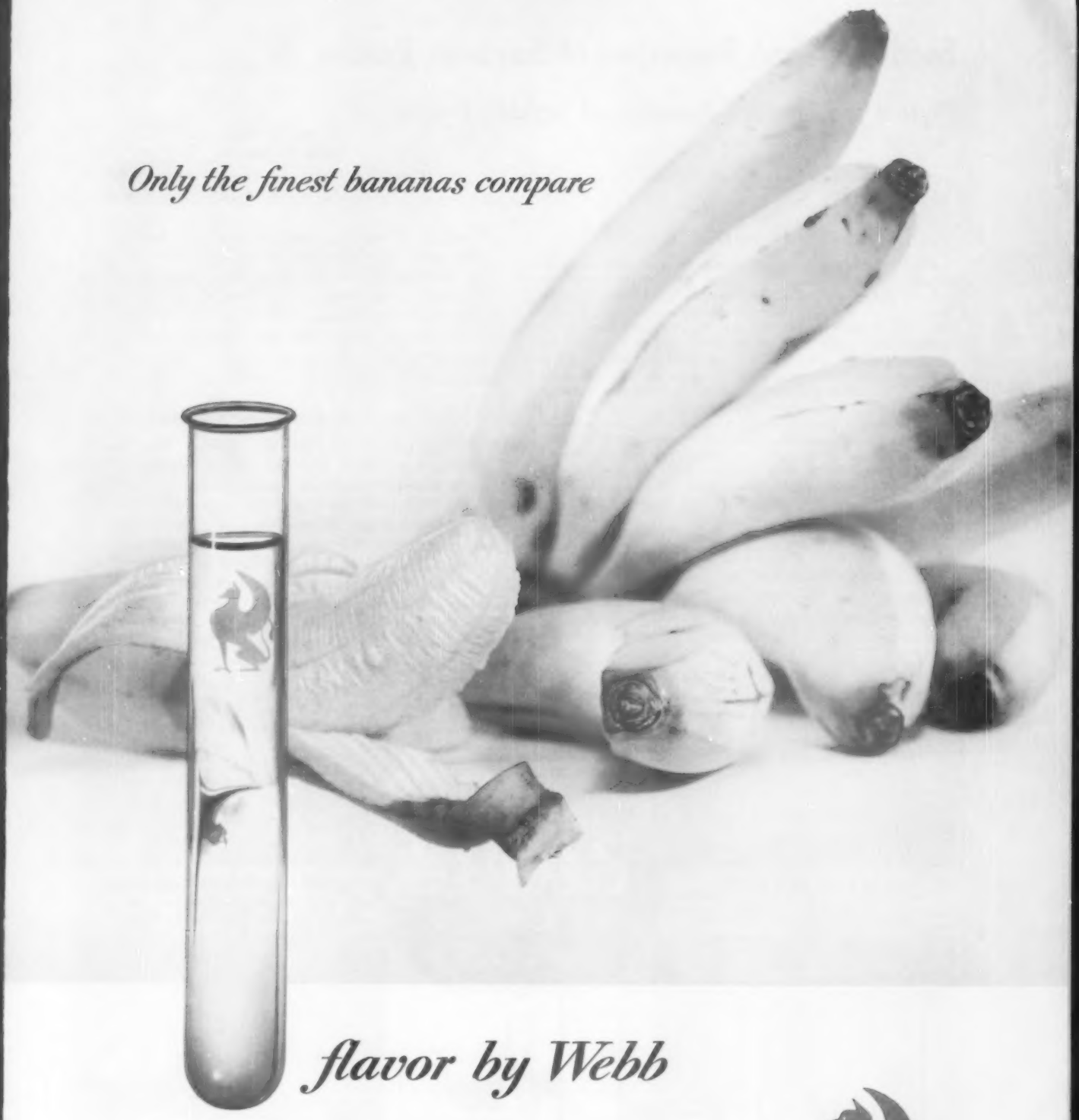

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Food Uses and Properties of Soybean Protein. II.

Physical and Chemical Properties of Soybean Protein[a]

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(Manuscript received June 28, 1960)

SUMMARY

Electrophoresis and ultracentrifugation show that soybeans contain a mixture of proteins. About 90% of the extractable proteins are globulins, and ultracentrifugal studies indicate four fractions having sedimentation constants of 2, 7, 11, and 15S. The 7S and 11S globulins have been partially purified and characterized. Little is known about the 2S and 15S fractions. The 7S globulin is dissociable into subunits, associates to higher molecular weight, and forms disulfide polymers. The 11S globulin has a molecular weight of about 350,000 and undergoes reactions similar to those of the 7S globulin. The 7S and 11S globulins are not interconvertible by association–dissociation reactions, which indicates that they are different proteins rather than aggregates of identical subunits. The whey fraction obtained in soybean protein isolation is a mixture of low-molecular-weight proteins. Two trypsin inhibitors, hemagglutinin, and three other fractions have been separated from whey protein by chromatographic methods.

PART I of this series (Smith and Wolf, 1961) reviewed the preparation of soybean meal and its protein fractions and their basic application to foods. Food utilization of soybeans in the U. S. is still so new that many problems must be solved before their main pattern of use is established. Part II reviews presently known basic physical and chemical properties of the protein of the soybean to provide a more scientific basis for solution of such problems.

The properties of soybean protein are still poorly understood, and there is confusion in the literature in certain areas. For example, in ultracentrifugal studies of soybean protein different sedimentation constants are reported for apparently the same components, and various workers interpret their results differently. We therefore attempt to show the relation between whole soybean protein and its various fractions, presenting a more unified picture of the number of components now believed present.

A review of the physical and chemical properties of soybean protein, to appear shortly (Bain *et al.*, in press), emphasizes industrial rather than food uses.

UNFRACTIONATED SOYBEAN PROTEIN

Briggs and Mann (1950) reported that electrophoretic analysis of a water extract containing 95% of the nitrogen of soybean meal indicated seven components or more, three of them accounting for about 75% of the total protein. Kondo *et al.* (1953) reported at least five electrophoretic components in a 1.5*N* sodium chloride extract containing 83% of soybean meal protein; two of the components account for about 80% of the total protein. Electrophoresis shows that soybean protein is a mixture, but does not clearly indicate the extent of heterogeneity, primarily because the components are poorly resolved under the conditions employed to date.

A clearer picture of the degree of heterogeneity is obtained by ultracentrifugation. Studies of unfractionated soybean protein at pH 7.6–7.8, 0.5 ionic strength, indicate four well-resolved fractions having approximate sedimentation constants of 2, 7, 11, and 15S (Naismith, 1955; Wolf and Briggs, 1956) (Fig. 1-a). Undoubtedly each of the four sedimenting peaks does not represent a single component (Wolf and Briggs, 1956) but hereinafter we refer to them as 2S, 7S, 11S, and 15S. Table 1 compares the sedimentation constants for unfractionated soybean protein as reported from two laboratories (Naismith, 1955; Wolf

[a] Presented at the Twentieth Annual Meeting of the Institute of Food Technologists, San Francisco, California, May 16–19, 1960.

[b] Laboratory of the Northern Utilization Research and Development Division, Agricultural Research Service, U. S. Department of Agriculture.

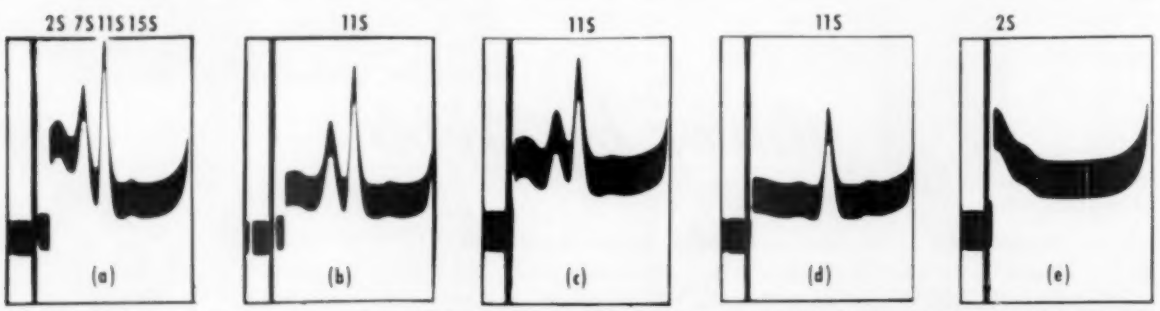


Fig. 1. Ultracentrifuge patterns for: (a) Water-extractable soybean proteins; (b) glycinin obtained by exhaustive dialysis of a solution of the proteins precipitated with 70% saturated ammonium sulfate from a salt extract of meal; (c) acid-precipitated proteins; (d) cold-insoluble fraction; and (e) whey proteins. Direction of sedimentation is from left to right, and sedimentation constants, in Svedberg units, are indicated in (a); in subsequent diagrams only the major peak is identified. Figures a, c, and e are directly comparable. Taken from Wolf and Briggs (1959).

Table 1. Sedimentation constants of soybean protein and its fractions.

Preparation	Ref.	$s_{20,w}$ values of sedimenting peaks			
		2S	7S	11S	15S
Unfractionated soybean protein					
10% NaCl-soluble protein	a	~2	6.6	10.6	15.0
Water-soluble protein	b	1.8	6.5	10.4	14.6
1*M* NaCl-soluble protein	b	1.9	6.7	10.6	14.2
0.5*M* $CaCl_2$-soluble protein	b	1.1	6.7	10.5	15.1
Soybean protein fractions					
Glycinin	c	....	7.97	13.06	....
Glycinin	a	~2	7.5	11.4	16.3
Glycinin	d	....	8.1	14.0	....
Glycinin	e	....	8.7	14.19	....
Acid-precipitated protein	a	~2	7.6	11.4	15.7
Cold-insoluble fraction	a	~2	7.4	11.8	16.9
Cold-insoluble fraction	f	~2	7.0	10.8	14.1
Purified 11S component	g	....	....	12.2	....
Albumin	d	1.98	....	....	....
Whey	h	1.75	6.13	....	....

[a] Naismith (1955). [b] Wolf and Briggs (1956). [c] Danielsson (1949). [d] Kretovich *et al.* (1956). [e] Kretovich *et al.* (1958a). [f] Briggs and Wolf (1957). [g] Wolf and Briggs (1959). [h] Rackis *et al.* (1959).

and Briggs, 1956). Agreement is excellent even though different solvents were used for extraction. Although the solvents differ in their ability to extract proteins (Smith *et al.*, 1938), ultracentrifuge patterns for soybean proteins extracted with them indicate no important differences in the relative amounts of each fraction.

Table 2 lists the amino acid composition of unfractionated soybean protein as recently reported from two laboratories (Kimura, 1956; Van Etten *et al.*, 1959) using the chromatographic procedure of Moore and Stein (1951).

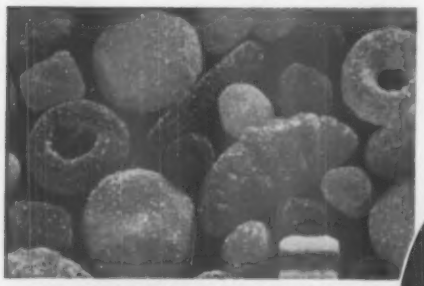
Table 2. Amino acid composition of soybean protein and its fractions.[a]

Amino acid	Unfractionated			Acid-precipitated	Heat-coagulated whey	
	Japanese[b]	Hawkeye[c]	Lincoln[d]	Lincoln	Hawkeye and Adams	Lincoln
Arginine	7.4	7.3	6.5	6.2	5.5	5.4
Histidine	2.6	2.6	2.6	2.1	2.6	2.6
Lysine	6.0	6.9	6.7	6.1	7.8	8.4
Tyrosine	3.9	3.8	4.4	3.9	5.6	4.8
Tryptophan	1.2	1.5	1.4	1.5	2.7	....
Phenylalanine	4.2	5.6	5.6	5.4	6.6	6.2
Cystine	0.80	1.4	1.4	1.2	1.7	....
Methionine	1.5	1.3	....	1.4	1.6	1.6
Serine	5.5	....	6.0	5.7	5.2	4.9
Threonine	4.7	3.9	3.9	3.6	4.7	4.5
Leucine	7.6	8.2	8.1	9.1	10.5	....
Isoleucine	6.2	....	5.3	....	6.6	....
Valine	5.3	....	5.6	5.4	7.3	7.1
Glutamic acid	19.3	20.1	18.5	20.8	14.7	....
Aspartic acid	14.0	11.9	11.3	12.0	13.5	....
Glycine	4.8	5.0	4.6	4.5	5.5	4.9
Alanine	5.0	4.2	4.0	3.9	5.3	5.1
Proline	4.4	....	5.3	5.6	5.8	....

[a] Data for Japanese variety from Kimura (1956); all other data from Van Etten *et al.* (1959). All data expressed as g/16 g N.
[b] Contained 14.62% N. Soybean variety was not indicated.
[c] Contained 16.1% N.
[d] Contained 15.8% N.

SOYBEAN PROTEIN FRACTIONS

Although reported sedimentation constants for whole soybean protein agree well, the constants for



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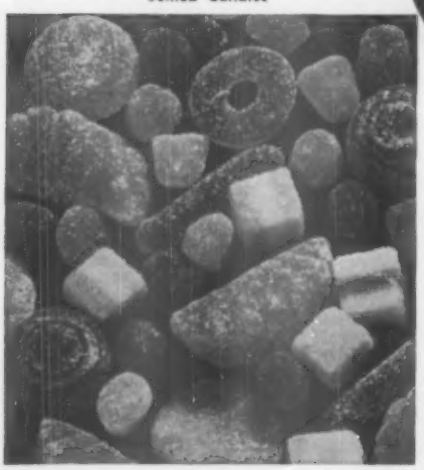

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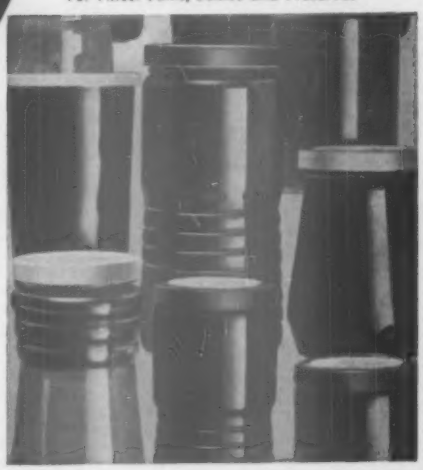

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components of the various soybean protein fractions do not (Table 1). Each component of the different fractions is classified as 2, 7, 11, or 15S to show its most probable relationship to the corresponding component(s) in unfractionated soybean protein. The wide variations for apparently the same components in various preparations may result from a) differences in protein concentrations and solvent systems, and b) use of dissimilar types of ultracentrifuges (Shulman, 1953; Taylor, 1952).

Glycinin. The classical fraction of soybean protein is glycinin, originally defined by Osborne and Campbell (1898) as the globulin obtained by salt extraction, ammonium sulfate precipitation and dialysis. The term glycinin, although sometimes used to describe other soybean protein fractions, is in this review restricted to Osborne's definition.

Heterogeneity of glycinin was first clearly indicated by Danielsson (1949), who found two ultracentrifugal components having $s_{20,w}$ values of 7.97 and 13.1S. The ultracentrifuge pattern, however, also suggests the presence of 2S and 15S components. Later, Naismith (1955) showed glycinin to consist of 2, 7, 11, and 15S fractions, with 11S protein present to the greatest extent. Although Kretovich *et al.* (1956) reported that glycinin consists only of the 7S and 11S components, present in equal amounts, careful examination of their schlieren patterns indicates the presence of 2S and 15S fractions. Those workers estimated the molecular weights of the 7S and 11S components to be respectively 138,000 and 336,000. They later (1958a, b) studied the ultracentrifugal behavior of glycinin and an ammonium sulfate fraction thereof under a wide range of pH values and ionic strength. They concluded that the protein consists of a monomeric 7S unit (designated S_8) that can dissociate into six identical subunits ($s_{20,w}$ = 2.4–3.1S) or can associate into a dimer and trimer respectively designated S_{11} and S_{14} components.

The discovery by Briggs and Wolf (1957) and Wolf (1956) that the 7S and 11S components are capable of forming disulfide-linked polymers makes the above ultracentrifugal studies difficult to interpret because the extent of polymerization in the glycinin preparations is unknown. Even when disulfide polymerization is eliminated, glycinin varies widely in ultracentrifugal composition, depending on the pH attained during dialysis (Wolf and Briggs, 1959). The isoelectric pH of the system is approached only after exhaustive dialysis, and then nearly 90% of the protein extracted from soybean meal with 1*M* sodium chloride is precipitated in the globulin fraction. Glycinin obtained by exhaustive dialysis (Fig. 1b) contained 2, 7, 11, and 15S components with the 7S and 11S proteins present in large amounts.

Heterogeneity of glycinin is also demonstrated by electrophoresis. The presence of two (Kretovich *et al.*, 1958c) and three components (Briggs and Mann, 1950; Kondo *et al.*, 1953) is reported, but there is no obvious correlation with the ultracentrifuge components.

Solubility of glycinin in water is increased by treatment with reducing agents such as cysteine (Kretovich *et al.*, 1958a; Kretovich and Smirnova, 1957). Insolubilization probably results from disulfide polymerization, which occurs during isolation, as is noted with the cold-insoluble fraction of soybean protein (Briggs and Wolf, 1957).

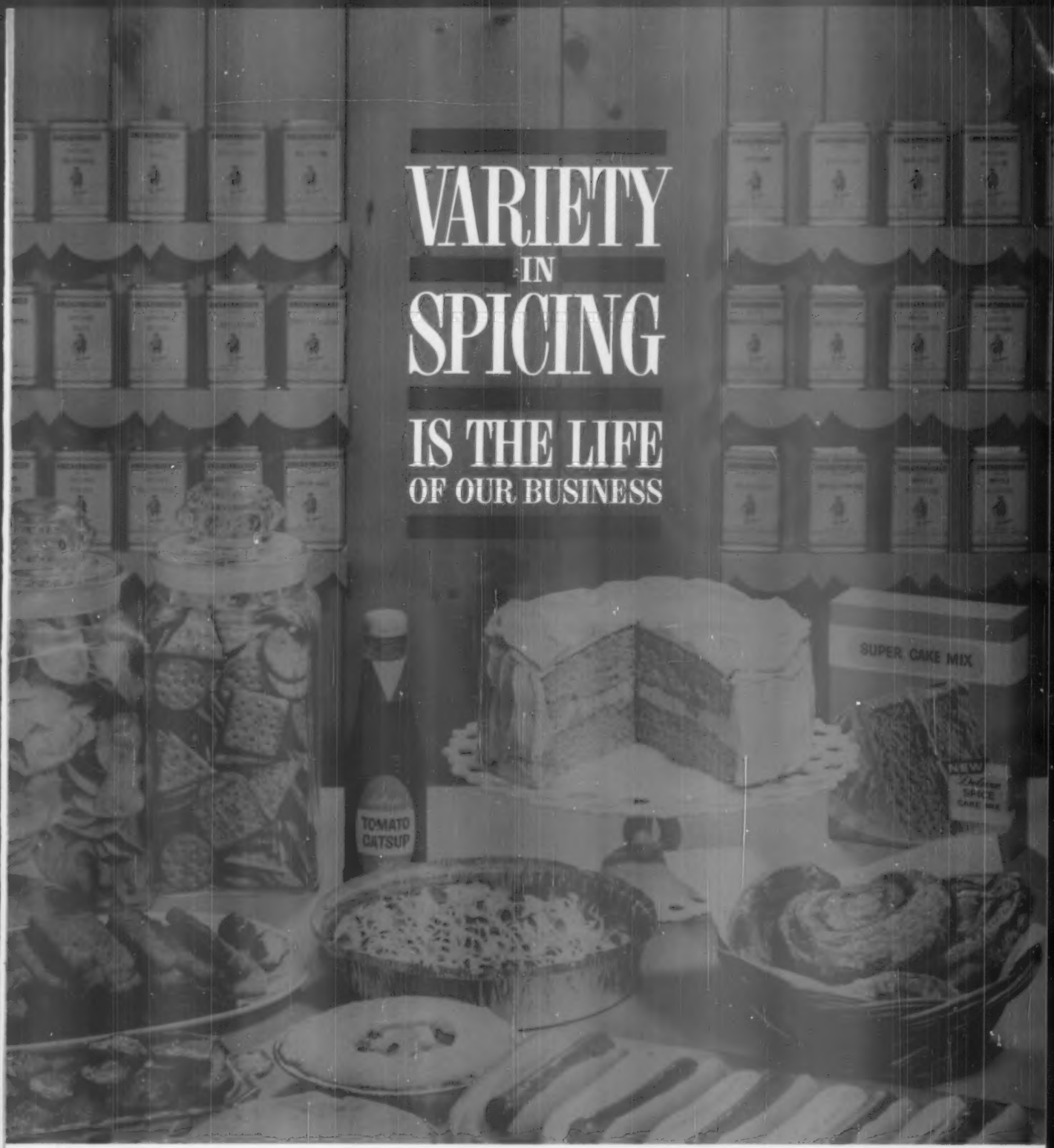
Beta-amylase, urease, and dehydrogenase activities are reported for glycinin; cysteine increases enzymatic activity under certain conditions (Kretovich and Smirnova, 1957). The finding of a multiplicity of enzyme activities, plus the known heterogeneity of glycinin, suggests contamination by individual enzymes rather than liberation of enzymatically active sites within the 7S and 11S molecules, as postulated by Kretovich and Smirnova (1957).

Acid-precipitated protein. Of primary interest to food technologists is the commercially available acid-precipitated fraction of soybean protein. Briggs and Mann (1950) investigated the electrophoretic behavior of acid-precipitated protein and observed resolution into at least four components. Smith *et al.* (1955), who got essentially the same electrophoretic patterns, reported the presence of one major and four minor components. One of their minor components appears to be a large aggregate removable from solution by high-speed centrifugation. Ultracentrifugal studies by Naismith (1955) showed at least four components corresponding to the four fractions observed in whole soybean protein. In confirming these results, Wolf and Briggs (1959) also showed that acidification of an aqueous extract of meal causes quantitative precipitation of the 11S and 15S fractions, nearly complete precipitation of the 7S components, and only 20–30% precipitation of the 2S fraction (compare Fig. 1-a, c, e).

An ultracentrifuge study of the acid-precipitated protein in acid solutions indicated a complex system very sensitive to pH and ionic strength suggesting the existence of association–dissociation reactions (Rackis *et al.*, 1957).

Comparison of ultracentrifuge patterns for glycinin and acid-precipitated protein (Fig. 1-b, c) shows only minor differences in relative distribution of components indicating that soybean globulins may be prepared simply by acidifying an aqueous extract of soybean meal. However, the acid-precipitated globulins are contaminated by nonprotein impurities (Smiley and Smith, 1946). Acid-precipitated protein prepared in the laboratory contains 0.3–1.6% ash and 0.5–1% phosphorus (McKinney *et al.*, 1949); commercial food-grade protein contains 2.2–2.5% ash (Circle and Johnson, 1958). The phosphorus impurities consist of phytate (McKinney *et al.*, 1949; Smith and Rackis, 1957), nucleic acid (Smith and Rackis, 1957), and possibly phosphatides (Smiley and Smith, 1946).

In Table 2 the amino acid composition of an acid-precipitated protein is compared with the unfractionated protein from a single soybean variety. The acid-precipitated protein has a slighly lower content of essential amino acids except for tryptophan and leucine, which are present in higher amounts.



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Cold-insoluble fraction and 11S protein. Cooling a concentrated aqueous extract of meal precipitates a cold-insoluble fraction of protein. This fraction is electrophoretically homogeneous but heterogeneous by the phase rule solubility test (Briggs and Mann, 1950). Heterogeneity was also observed by ultracentrifugation, which indicated the presence of 2, 7, 11, and 15S components (Fig. 1-d), with the 11S protein as the major component (Briggs and Wolf, 1957; Naismith, 1955). Cold precipitation from a water extract is highly selective for separation of the 11S component; the cold-insoluble protein may be regarded as a subfraction of the soybean globulins (compare Fig. 1-b, d).

A partially purified preparation of the 11S protein is also obtained by ammonium sulfate precipitation as described by Naismith (1955). A study of the effects of pH and ionic strength on this preparation revealed that at 0.1 ionic strength in the pH range of 6.9–9.5 the 11S protein associates to form an ill-defined faster-sedimenting component. A more detailed study confirmed Naismith's work and supplied additional information (Briggs and Wolf, 1957; Wolf, 1956; Wolf and Briggs, 1958). One previously mentioned property of the 11S protein (isolated as the cold-insoluble fraction) is its ability to form disulfide-linked polymers under the relatively mild conditions of precipitation by cooling, dialysis, or acidification. Recent reports (Deutsch and Morton, 1957; Durrell and Cantoni, 1959) indicate that the reaction is not unique to the 11S protein. Depolymerization of its disulfide polymers is possible with a wide variety of disulfide-cleaving reagents, including mercaptoethanol, sulfite ion, and sodium borohydride. Depolymerization increases its solubility and reduces the turbidity of its solutions. Sulfhydryl-blocking reagents, such as iodoacetamide and *N*-ethylmaleimide, prevent re-polymerization of the monomer of the 11S component.

Another property of the 11S protein is its ability to undergo conformation changes probably consisting of dissociation into subunits about one-eighth the size of the 11S molecule (Walf and Briggs, 1958). These changes occur at low ionic strength at alkaline pH values and at moderate ionic strengths in acid solutions, in the presence of the detergent sodium octylbenzene sulfonate, and in the presence of low concentrations of urea. The effects of pH and ionic strength suggest that dissociation involves rupture of weak secondary forces, such as hydrogen bonds and van der Waal's interactions, by electrostatic repulsion between positively or negatively charged subunits (Wolf and Briggs, 1958; Wolf *et al.*, 1958a) Acid dissociation is not a freely reversible process, presumably because of configurational changes in the subunits (Wolf *et al.*, 1958b).

An 11S protein of about 90% purity was obtained by extraction of the acid-precipitated protein with sodium chloride solution of graded ionic strength (Wolf and Briggs, 1959). Molecular-weight determinations yielded values of 363,000 and 345,000 respectively by sedimentation-diffusion and light scattering.

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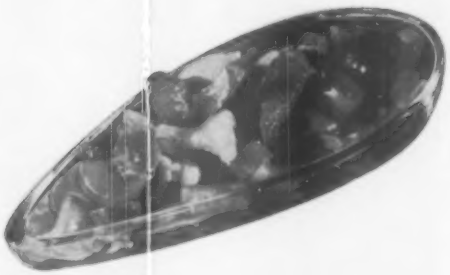

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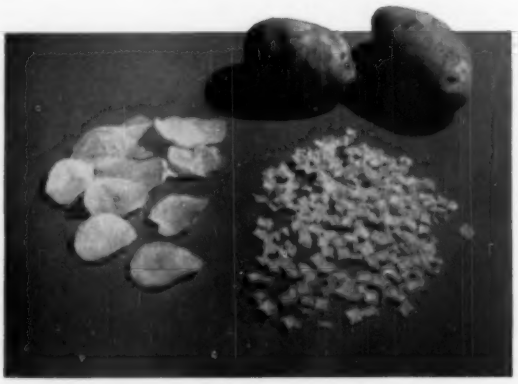

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These values are on the same order as that reported by Kretovich, 380,000, for an 11S preparation obtained by ammonium sulfate fractionation (Kretovich *et al.*, 1958a).

Ultracentrifuge patterns for glycinin and acid-precipitated protein (Fig. 1-b, c) are quite similar, whereas the pattern for the cold-insoluble fraction (Fig. 1-d) differs considerably because of its low content of 2S and 7S components. In addition, there are chemical differences. For example, the nitrogen content of glycinin is 16.4–16.6% (Kretovich *et al.*, 1956; Wolf and Briggs, 1959) and for the cold-insoluble fraction, 17.1–17.3% (Briggs and Wolf, 1957; Wolf *et al.*, 1958a). All three fractions have 11 *N*-terminal amino acids in common (Campbell *et al.*, 1957). The two globulin preparations are quite similar, as expected on the basis of ultracentrifugal studies, but both differed significantly from the cold-insoluble fraction in the amounts of major *N*-terminal amino acid residues present (Campbell *et al.*, 1957). Since the greatest difference between the globulin preparations and the cold-insoluble fraction is in the relative concentration of 7S and 11S components, the *N*-terminal studies suggest the presence of different polypeptide chains in the 7S and 11S proteins.

N-terminal studies of the cold-insoluble fraction by Campbell *et al.* (1957) also indicate that the 11S protein contains a minimum of four polypeptide chains and that three of these chains may be different. If one polypeptide chain per subunit, and equal chain size are assumed, the 11S protein of 350,000 molecular weight (Wolf and Briggs, 1959) consists of four subunits of about 90,000 molecular weight, compared to a molecular weight of about 20,000 reported by Kretovich *et al.* (1958b) for the subunits of glycinin. However, Campbell's value is a minimum uncorrected for contamination of the 11S protein by the 2, 7, and 15S components also present in the cold-insoluble fraction; Kretovich's value is an average obtained for the mixture of subunits obtained upon dissociation of the 7, 11, and 15S components of glycinin.

7S Protein. The 7S ultracentrifugal peak in unfractionated soybean protein is a composite. At least two of the minor proteins of soybean have sedimentation constants in the range of 5–7S (Wolf and Briggs, 1956). The major portion, however, has the properties of a globulin and occurs in glycinin and in acid-precipitated protein; crude preparations of this 7S globulin obtained by dissimilar methods have similar properties, suggesting that it is a single component.

A 7S preparation obtained by ammonium sulfate precipitation indicated that at 0.1–0.2 ionic strength the 7S protein is converted into a sedimenting species of 10.5S, probably a dimer that is resolvable from the 11S component upon ultracentrifugation (Naismith, 1955). This behavior was confirmed with a 7S fraction isolated from acid-precipitated protein by varying ionic strength and temperature (Wolf, 1956), and association of the 7S component into the dimer was observed at pH 9.0, 0.01 ionic strength. This reaction represents a distinct difference between the 7S and 11S proteins. Under these conditions the 11S protein

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is about 50% dissociated into subunits (Wolf and Briggs, 1958); however, like the 11S the 7S protein is also broken into smaller units when treated with the detergent sodium octyl benzene sulfonate; and it is also able to form disulfide-linked polymers (Wolf, 1956). Disulfide polymers of the 7S and 11S components apparently exist, preformed, in hexane-extracted meal (Wolf, 1956; Briggs and Wolf, 1957), but their existence in the whole bean is still unknown.

The depolymerizing action of cysteine and other disulfide-cleaving reagents on polymers on 7S and 11S proteins probably explains both the decrease in viscosity noted when solutions of unmodified acid-precipitated protein are treated with these reagents (Circle and Johnson, 1958; Hall, 1955) and the increase in solubility noted when glycinin is treated with cysteine or glutathione (Kretovich and Smirnova, 1957; Kretovich *et al.*, 1958a). In an ultracentrifuge study of the latter effect, Kretovich *et al.* (1956) found that when glycinin was dissolved in neutralized cysteine hydrochloric (pH 7, ionic strength estimated at 0.02), the solution contained three sedimenting components, of $s_{20,w}$ values 7.5, 10.7, and 13.5S. The 10.7S peak, which was abolished by addition of sodium chloride, is probably the dimer of the 7S protein formed at low ionic strength and dissociated at high ionic strength (Naismith, 1955), rather than the product resulting from treatment of the 7S protein with cysteine.

Soybean whey proteins. Whey is obtained either by removal of the acid-precipitated proteins in a water extract or by extraction of the meal at pH 4.5. Such a solution contains about 30% of the meal. Whey contains an unknown number of albumins and globulins, water-soluble carbohydrates (MacMasters *et al.*, 1941), nonprotein nitrogen (Becker *et al.*, 1940), salts, vitamins, phytate (Smith and Rackis, 1957), and other high- and low-molecular-weight components. Processors of isolated protein are concerned about the whey solution because it presents a waste disposal problem.

Biologically active proteins reported in the whey fraction are trypsin inhibitor (Kunitz, 1946), β-amylase (Rackis *et al.*, 1959), soybean hemagglutinin (Liener and Hill, 1953; Wada *et al.*, 1958), phosphatase (Mayer *et al.*, unpublished), lipoxidase (Theorell *et al.*, 1947), phytase (Fontaine *et al.*, 1946), and others (Circle, 1950). The most notable work on the whey protein has been the isolation in highly purified states of crystalline lipoxidase by Theorell *et al.* (1947); the crystalline soybean trypsin inhibitor (STI) by Kunitz (1946); and hemagglutinin by Wada *et al.* (1958).

When Ham *et al.* (1945) discovered STI they reported it an anti-growth factor, but further research (Borchers and Ackerson, 1950) indicates that antitryptic activity in other legumes does not inhibit growth. Later reports (Ribelin *et al.*, 1960) show that feeding STI enlarges the pancreas to approximately double its normal weight. Immediately after ingestion of STI, intestinal tryptic activity is low but increases steadily and at the end of 6 hours is three times nor-

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mal (Lyman, 1957). Secretion of other pancreatic enzymes also increases. Liener's hemagglutinin (Liener and Hill, 1953) appears to depress the appetite; whether restricted food intake accounts entirely for the slower rate of growth or whether part is from a toxic effect of the hemagglutinin is not yet known.

The number of proteins in whey and their similarity in size and charge pose a difficult problem in separation. Wolf and Briggs (1959) found only two resolvable peaks in the ultracentrifuge pattern corresponding to components with $s_{20,w}$ values of about 2S and 6S, with the former in the higher concentration. Smith *et al.* (1955) found five electrophoretically distinct components. Chromatographic and biochemical studies show a *much* greater number of components.

Rackis *et al.* (1959) found that the most effective method of separating whey proteins is by column chromatography using diethylaminoethyl cellulose (DEAE-cellulose). Using a gradient elution procedure they obtained 13 chromatographic peaks, indicating a minimum of 13 proteins. The various peaks were not separated well enough for further characterization, and a stepwise elution procedure was developed that divided the whey protein into six fractions. These were then characterized by electrophoresis and ultracentrifugation. Their biological activity is shown in Table 3.

Table 3. Analysis of protein fractions isolated from soybean whey.

Fraction	Electrophoretic data		Ultracentrifuge data		Biological assay specific activity values[b]			
		Mobility[a] $\times 10^{-5}$	$s_{20,w}$ values $\times 10^{-13}$	Total area, %	Phosphatase	β-amylase	SBH[c]	STI[d]
I	A	0.08	0.99	84	1.2	0	868	0
	B	0.55	5.85	16				
	C	1.13						
II	D	2.70	2.85	21	24	0	9123	0
			6.14	79				
III	E	3.56	1.58	36	43	3.6	1595	0
	F	3.89	5.66	64				
	G	4.37						
IV	H	5.0	1.73	73	14	2.4	66	0.45
	I	5.45	5.64	27				
	J	5.99						
V	K	6.55	2.29	87	2	1.7	100	1.28[e]
	L	7.17	5.62	13				
VI	M	7.86	1.75	100	0	0	23	1.04[e]
Whole whey	1	6.30						
	2	7.56						
	3	9.96						
	4	3.11	1.75	59				
	5	4.72	6.13	41	20	2.9	1480	0.387

[a] Mobilities as measured on the ascending boundaries.
[b] Values calculated on the basis of 1 mg of protein (N × 6.14).
[c] SBH, Soybean hemagglutinin.
[d] STI, Soybean trypsin inhibitor.

No biological activity occurred in the first fraction; the second was mostly hemagglutinin. The sixth fraction corresponded to Kunitz' trypsin inhibitor; the fifth is about 85% of a second trypsin inhibitor, with a somewhat lower sedimentation constant, lower electrophoretic mobility, and higher activity than the Kunitz inhibitor.

Amino acid composition of the heat-coagulable whey proteins is given in Table 2.

General Conclusion

Soybean proteins constitute a very complex biological system that calls upon all the skill and ingenuity of the protein chemist to separate and characterize the individual components. At present it appears that the 7S and 11S components observed in acid-precipitated soybean protein or glycinin fraction are distinct proteins that account for about 70% of the total soybean protein. This conclusion differs from that of Kretovich and coworkers (Kretovich *et al.*, 1958 a, b; Smirnova *et al.*, 1959). They reported that the globulin fraction consists of identical subunits (MW 20,000) existing in various states of association, depending on pH and ionic strength. Their view appears incorrect for the following reasons: a) Stable fractions containing widely varying ratios of the 7S and 11S components are obtained by a number of fractionation procedures (Naismith, 1955; Wolf and Briggs, 1959); b) partially purified 7S and 11S proteins are not interconvertible and their physical properties are distinctly different (Briggs and Wolf, 1957; Naismith, 1955; Wolf, 1956; Wolf and Briggs, 1958); *N*-terminal amino acid analysis indicates that there are in the 11S component at least three chemically different subunits that may also differ from the subunits of the 7S component (Campbell *et al.*, 1957). Although the 7S and 11S globulins are different proteins they undergo similar types of reactions, but not necessarily under identical conditions. At least three such reactions are now known: a) Dissociation into subunits (Wolf, 1956; Wolf and Briggs, 1958); b) association into noncovalent-linked polymers (Naismith, 1955; Wolf, 1956); and c) formation of disulfide-linked polymers (Briggs and Wolf, 1957; Wolf, 1956). The importance of these reactions in food products is unknown.

Establishing that soybean protein consists of distinct components emphasizes the major problem, and perhaps the most difficult one confronting the investigator studying soybean proteins: isolation of these components in a pure state. Because of the complexity and multiplicity of reactions of the various components, studies to date, particularly on crude fractions such as glycinin and acid-precipitated protein, are of limited usefulness and serve only as rough guides for future work. Perhaps chromatographic techniques will be as useful for purifying the 7S and 11S components as they have been for unraveling the complex whey protein system.

Choice of solvent is important in using ultracentrifugation for studying soybean protein fractionation. At present, a buffer of high ionic strength, near neutrality and containing a reducing agent, is most suitable. Used routinely in our laboratory is a phosphate–sodium chloride buffer, pH 7.6, 0.5 ionic strength containing 0.01*M* mercaptoethanol (Wolf and Briggs, 1959). This buffer system gives the simplest ultracentrifuge pattern because association–dissociation reactions of the 7S and 11S components are suppressed by the high ionic strength and because disulfide polymerization of the same proteins is prevented by the mercaptoethanol. This solvent is also

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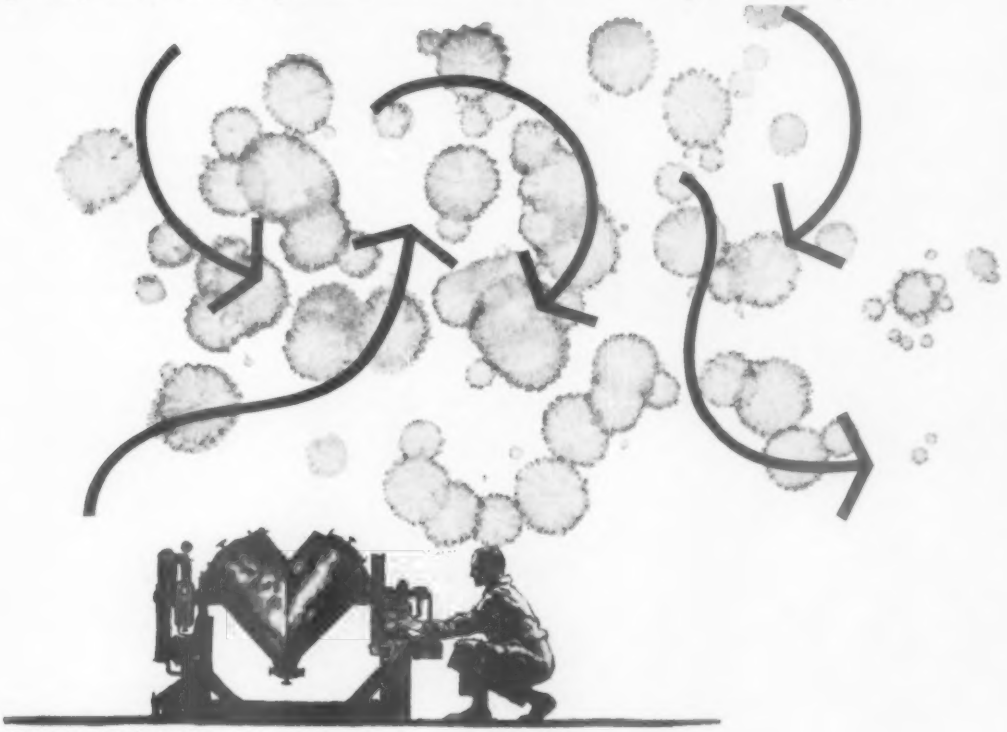

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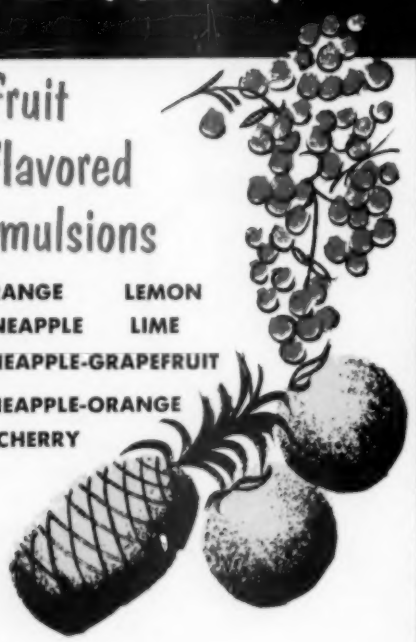
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recommended as reference state when ultracentrifuga- is used to determine whether a given treatment has caused denaturation of the 7S and 11S proteins.

When the 7S and 11S globulins are studied in the presence of salts, they consist of aggregates or association products of subunits. Whether they exist in the soybean in the subunit state or 7 and 11S state is unknown. However, ultracentrifugal examination of a water extract of meal containing only the salts present in soybeans showed no evidence of protein in the subunit state (Wolf, 1956). The 7S protein appeared to be in the dimer state and the 11S protein was partially associated, similar to the behavior of these components at pH 7.8, 0.1 ionic strength (Naismith, 1955). The time factor in this experiment was held at a minimum by extracting for only 5 minutes and by analyzing in the ultracentrifuge immediately after removal of the insoluble portion of the meal. If the proteins exist in the subunit state in the meal, they must be highly reactive and the 7S and 11S states must form very rapidly. Because the 7S and 11S components are stable under mild conditions, they may be regarded as possessing a quaternary structure in addition to the usual primary, secondary, and tertiary structure of proteins (Bernal, 1958). Based on this premise the logical approach to ultimate characterization of the major components of soybean protein consists in purification of the 7S and 11S components followed by dissociation, separation, and characterization of the individual subunits.

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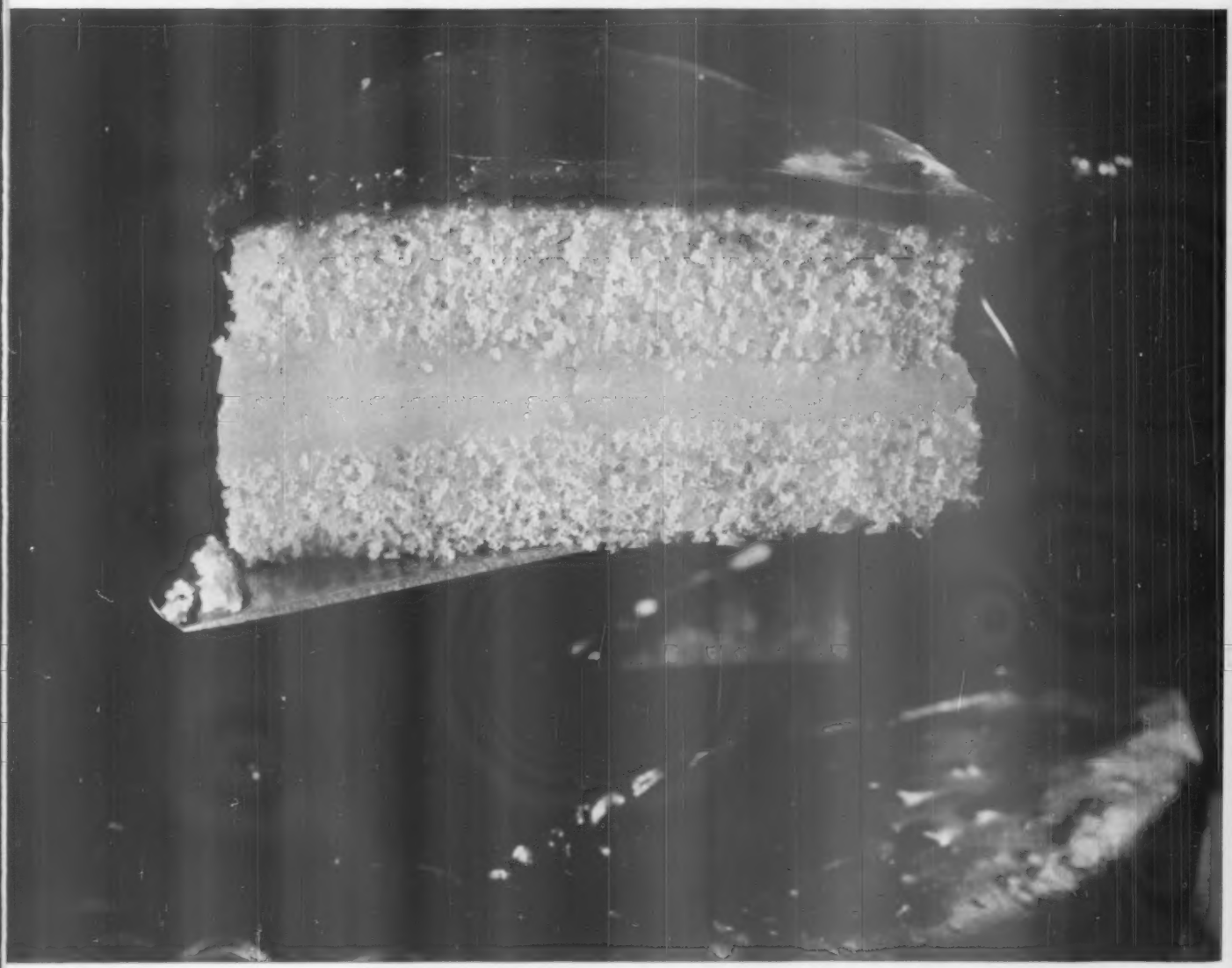

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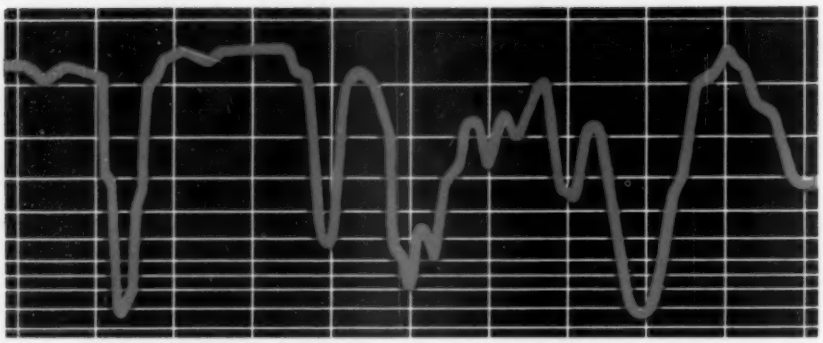

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Use of the Kramer Shear-Press in Forecasting Harvest Dates for Fordhook Lima Beans

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(Manuscript received October 31, 1960)

SUMMARY

The shear-press can be helpful to the field man in establishing harvest dates for Fordhook limas. With careful representative sampling and use under specific conditions, shear-press values supply useful, added information on the relative growth rates of the various fields, particularly in the all-green stage, a few days before harvest. The shear-press will not, however, pinpoint a desired pale count for a given day.

IN TODAY'S competitive frozen-food market, the problem of harvest time for Fordhook lima beans is economically important. With buyers demanding a nearly all-green product, an excess of pale green beans (pales) means lower profits to the packer, while the grower is interested in harvesting as large a tonnage as possible under the conditions of his contract with little or no dockage because of excess pales and whites. The field man has the task of satisfactorily compromising these divergent views.

Various instruments and procedures have been suggested and tried for rapid and accurate prediction of optimum maturity and harvest dates for lima beans (Culpepper and Caldwell, 1945; Culpepper and Magruder, 1943; Kramer and Hart, 1954; Kramer *et al.*, 1951; Kramer and Mahoney, 1940; Lee, 1943, 1948; Salunkhe and Pollard, 1955; Sterling, 1955a, b). To date, the main guides used by most field men for the Fordhook variety are the number of growing days in the ground or the days from full bloom, the number of full pods per vine, and the percentage of pales. Until the beans begin changing color, however, the field man has little to guide him on relative rates of maturity development in various fields. A rapid objective method giving this kind of information, from about a week to ten days before expected harvest, would be helpful—particularly under conditions of hot weather with several fields appearing to be about equally mature.

In 1951, Kramer *et al.* reported the development of the multipurpose shear-press. This instrument provides rapid measurement of such quality factors as hardness, succulence, and/or stringiness for various raw and processed foods. With lima beans, the shear-press forces a series of metal grids vertically through a standard volume or weight of beans in a rectangular slotted box. The pressure necessary to shear through raw beans reflects their maturity. Increasing maturity results in harder beans, requiring greater pressure.

Several reports (Kramer, 1960; Kramer and Hart, 1954; Kramer *et al.*, 1951) have indicated the usefulness of the shear-press in predicting the quality of raw and processed lima beans and in estimating harvest dates and yields for baby lima bean varieties. In Utah, Salunkhe *et al.* (1959) reported no significant effect of harvest time on shear-press values of several varieties of lima beans, including Fordhooks, when dry and shriveled beans were removed from the samples tested. When dry and shriveled beans were left in the samples, however, shear-press value generally increased with each succeeding harvest.

In California, where most of the Fordhooks for freezing are grown, no published shear-press data are available. The present studies were conducted to determine the usefulness of the shear-press as an aid in forecasting harvest dates of Fordhook limas grown under the more typical conditions found in California.

Comparisons were made among shear-press values of raw Fordhooks, the percentage of pales by count or weight, and the alcohol-insoluble solids (AIS) of some raw bean samples at harvest and during a 7–10-day period before expected harvest. The per cent AIS is generally used to reflect the physiological maturity of starchy vegetables such as peas and limas.

PROCEDURE

All data were collected under field and plant operating conditions. During the 1956 and 1957 seasons, the shear-press was used at the processing plant of Santa Clara Frosted Foods Co., Santa Clara, California, and the 1958 and 1959 data were obtained at the vining station of Spiegl Farms, Inc., at Oxnard, California.

During the 1956 and 1957 seasons, most of the data were collected on harvested raw Fordhooks of the Concentrate variety. Three shear-press readings were made on each lot. Per cent color by count (250–300 beans per lot) was determined by the USDA inspection procedure for color grading of frozen Fordhook lima beans. Before color grading, the beans were placed in boiling water for 3 min and then immediately cooled in running tap water. The limited preharvest data collected at Santa Clara were from vines in specific rows in several areas of the field. Beans were hand-shelled at the plant, and generally only one shear-press determination was made, because of the limited material available.

A more detailed preharvest testing program was followed in 1958 and 1959 at the Oxnard vining station. Fields of Regular Fordhooks were systematically sampled by dividing each field into quarters. Each quarter was sampled separately so that the path followed was roughly an arc of each quarter. Pods were removed by hand and hulled in a laboratory podder. In 1958, beans from the four sections of the field were mixed together for the various determinations; in 1959, the beans from each quarter were assayed separately. Generally 10–15 vines were collected from each quarter of a field. Three shear-

[a] A laboratory of the Western Utilization Research and Development Division, Agricultural Research Service, U. S. Department of Agriculture.

[b] Spiegl Farms, Inc., Salinas, California.

[c] Agricultural Marketing Service, Fruit and Vegetable Division, Processed Products, Standardization and Inspection Branch, U. S. Department of Agriculture, San Jose, California.

Table 1. Coefficients of correlation: (a) shear-press with percentage of pales, (b) alcohol-insoluble solids with pales, and (c) shear-press with alcohol-insoluble solids.

Year	Fordhook variety (area)	Color method	Shear-press with pales (%)[a]	AIS (%) with pales (%)[a]	Shear-press with AIS (%)
1956	Concentrate (Santa Clara, Calif.)	By count	+.72 (61)[b]	+.35(10)[b]	+.94 (11)[b]
1957	Concentrate (Santa Clara, Calif.)	By count	+.76 (14)	+.66 (14)	+.91 (14)
1958	Regular (Oxnard, Calif.)	By weight	+.65 (127)	+.53 (54)	+.91 (108)
1959	Regular (Oxnard, Calif.)	By weight	+.62 (115)	.....[c]	+.93 (10)

[a] Whites included.
[b] Numbers of samples shown in parentheses.
[c] Insufficient data.

press readings were made on each lot, using 160 g per sample. The rate of travel of the shear-press power piston for all determinations was 4 sec per inch. Per cent color by weight was determined by the same procedure as for the per cent color by count, except that 1 lb of beans for each preharvest lot and 50 oz for each harvest lot were color graded. If sufficient amount remained, 1–2 lb of the raw limas were placed in a polyethylene bag, tied with a rubber band, and held 6–12 hr in chipped ice until transfer to 0°F storage for later AIS analyses by the method described by Sterling and Boggs (1954).

RESULTS AND DISCUSSION

Table 1 presents correlation coefficients for shear-press values, percentages of pales, and percentages of alcohol-insoluble solids. The correlations of color (per cent pales) with shear-press values and with AIS are low. The correlations of shear-press with AIS, however, have been consistently high, ranging from +.91 to +.94. The shear-press vs AIS correlation data cover a greater maturity range because of inclusion of the all-green samples, as well as those having a pale count. These data indicate that percentage of pale beans is not a reliable guide to Fordhook maturity, but that the shear-press values generally reflect bean maturity.

These correlations are based on both preharvest samples plus harvest samples. The harvest samples were collected at the viner station after vining and passing through the Clipper Cleaner, where the small beans dropped through a screen with holes of ½ in. diameter. Whites and shrivels, when present, were left in the sample. Correlations between shear-press and color are not improved, however, when the data are divided into preharvest and harvest lots, and lots without white beans (Table 2).

Table 2. Coefficients of correlation of shear-press with pale greens (%) for preharvest and harvest lots and lots without white beans (1959).

Lots	Number of lots	Correlation coefficient (shear-press with pales)
Preharvest containing no whites	55	+.57
Preharvest, total	60	+.48
Harvest containing no whites	39	+.62
Harvest, total	55	+.73
Total, preharvest and harvest	115	+.62

The generally poor correlations between Fordhook color and both shear-press and AIS point up the fact that pale Fordhooks may be immature and tender, or that green limas may sometimes be relatively mature and hard, probably reflecting genetic, cultural, or climatic influence. Lee (1943) noted this same irregularity with baby lima beans that normally turn white with maturation. The difficulty sometimes encountered in color-separating Fordhooks by the usual brine density method would also appear to be related to these color-maturity abnormalities.

In spite of the poor relationship of shear-press to color, and the fact that grade is based primarily on color, the field men who have used the shear-press have found it helpful in establishing harvest dates. This is probably best explained by again noting that physiological maturity, expressed as percentage of AIS, is reflected by the shear-press. Thus the field man, having followed a periodic, systematic sampling procedure from several days before expected harvest, in the all-green stage, and up to harvest, has this additional information on relative rates of maturity development of various fields and can better determine in what order to harvest.

Fig. 1 shows some typical growth curves for several 1958 fields planted consecutively. The shear-press range is shown over which pales first began to appear (about 850–1050 lb pressure), and another range (1050–1125 lb pressure) where the desired pale count (4–6%) for harvest occurred.

The shear-press can sometimes be helpful in predicting a sudden jump in pale count when an all-

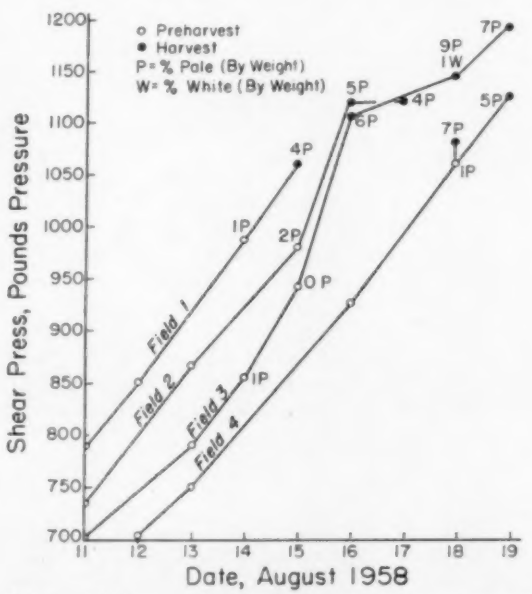

Fig. 1. Relation of shear-press values to time and color development for 1958 fields.

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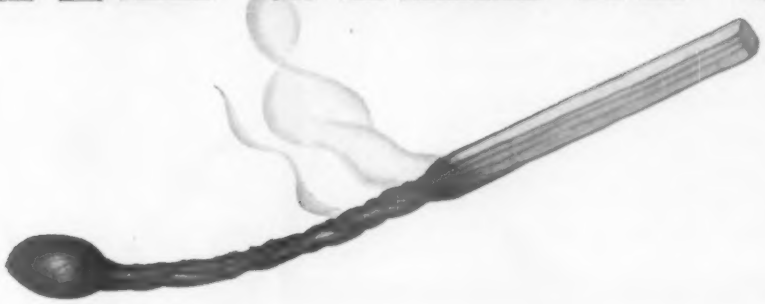

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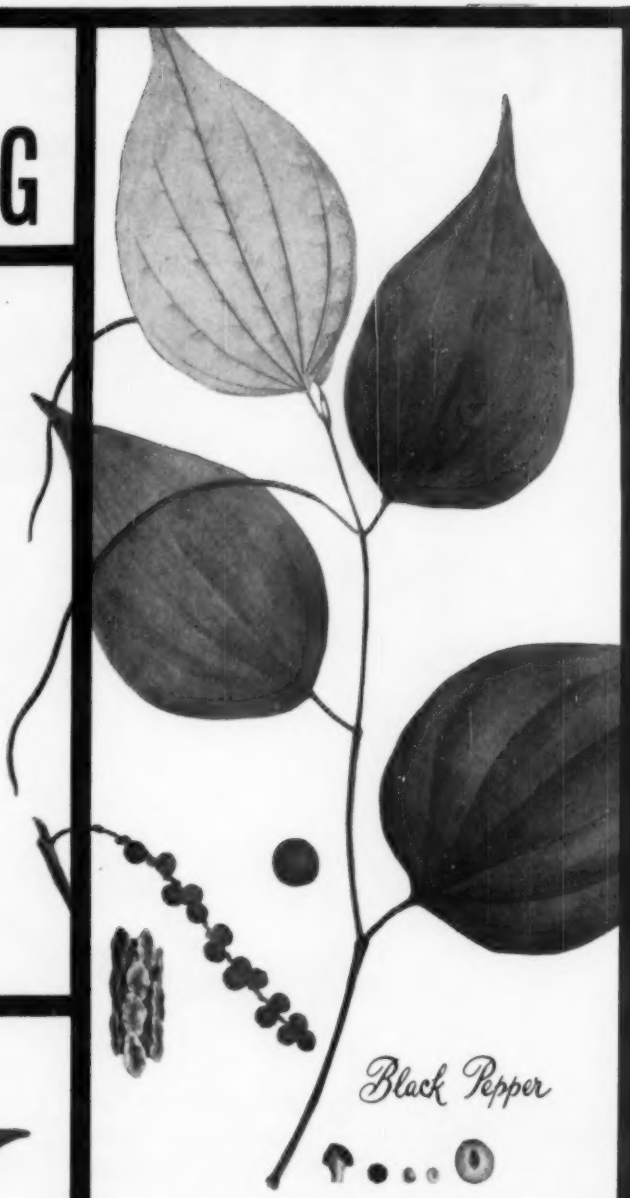

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green field has a shear-press value in the range of other harvested fields having the optimum 4–6% pales. For example, the morning preharvest sample of field 4 (Fig. 1) indicated 1% pale at a shear-press value of 1060, whereas harvest samples later the same day and the following day respectively showed 7 and 5% pales.

Fig. 2 shows growth curves for the first fields harvested during the 1959 season. Some of the shear-press values for fields 5 and 6 that appear out of line are most likely due to sampling errors. Other rapid changes, however, were not predictable, such as the one-day jump of almost 200 shear-press units in field 8, instead of the approximately 50 units per day expected.

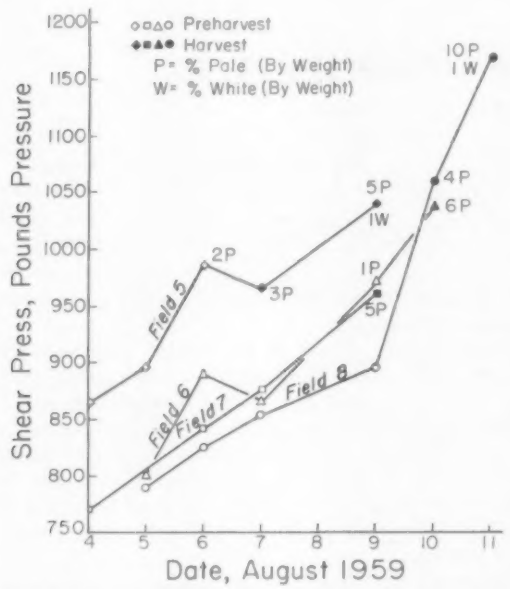

Fig. 2. Relation of shear-press values to time and color development for 1959 fields.

These curve irregularities emphasize the importance of adequate careful sampling procedures in order to minimize false or misleading shear-press values and to evaluate these growth curves more accurately. However, some of the erratic points on these growth curves and some of these poor shear-press and color relationships are better understood by the field man. Being "on the spot," the field man will be aware of such factors as irregular sets, "hot spots," time of day samples were pulled, soil moisture conditions, borderline green-pale beans, vines with beans turning prematurely pale, and various other factors that can influence this color and shear-press relationship—and would be considered in judging when a field is ready for harvest.

The optimum 5% pales at harvest occurred in these early 1959 fields at a slightly lower shear-press value than was found in 1958, and the 1959 fields harvested toward the end of the season were about 50–100 shear-press units below the early 1959 fields for the same percentage of pale green beans. Thus the proper application of the shear-press data in forecasting Fordhook harvest dates is to some extent an art, acquired from experience.

Fields are generally harvested in the order of planting. In some cases, however, this order is not usable. Plotting shear-press values against time can alert the field man to this situation several days before harvest. Fields 9 and 10 (Fig. 3) illustrate this point.

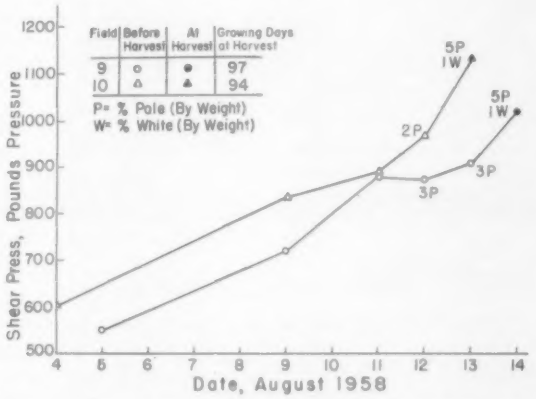

Fig. 3. Comparison of shear-press growth curves and color development between two fields planted three days apart.

Field 10, with three fewer growing days, was harvested a day before Field 9, and both fields had a reasonable pale count at harvest. Without the advance warning given by these plotted shear-press values, however, Field 10 might have been harvested a day or so later, with considerable increase in pales and whites.

Acknowledgment

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Thermal Properties of Foods at Low Temperatures. I.

Specific Heat

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(Manuscript received November 10, 1960)

SUMMARY

A rapid method is described for continuous determination of the specific heat of foods over a wide range of temperatures below the freezing point. With this method, the specific heat of fats, gelatin, and water were measured. From these data, it is possible to estimate the specific heat of foods, based on their composition, using an appropriate correction factor. The possibility of vitreous ice formation is suggested to explain the discrepancies between the observed and computed specific heat values for meats and fish.

LIQUID NITROGEN has been used successfully in maintaining temperatures below 0°F during shipment of frozen foods. Investigations on cooling foodstuffs with liquid nitrogen indicate no harm to the product (Moiseeva and Piskarev, 1959; and Moline, 1959). Foods tested include a large variety of frozen meats, vegetables, fish, and orange juice.

It was then necessary to measure the values of the specific heat of foodstuffs in order to determine accurately the volume of liquid nitrogen required to cool a given amount of food to a given temperature. Knowledge of the specific heat of a sample is also required for calculating cooling and thawing times.

LITERATURE REVIEW

Before 1942, most available information on the specific heat of foodstuffs was based on the work of Siebel (1892). He reasoned that since foods were composed of water and solid matter, it should be possible to calculate their specific heat above the freezing point by adding the product of the weight fraction of water present and its specific heat to the product of the weight fraction of the solid matter, and 0.2 as its specific heat. Below the freezing point he used only the specific heat of ice.

More recently, Short *et al.* (1942) measured the specific heats of some fruits, vegetables, meats, and sugar solutions, and found that the assumptions of Siebel were not correct, especially in the region between −15°C (+5°F) and +4.4°C (+40°F). In the region below −15°C, however, Short states that the numerical values are roughly the same as would be obtained by averaging the specific heat of ice and the solid matter.

The work of Short and co-workers was extended (Staph, 1949) in order to determine what relationship, if any, existed between heat capacity and the composition of foodstuffs. The lower temperature limit of his bomb calorimeter was −40°C. The foods measured were fruits and vegetables. He was unable to measure any food containing more than 3% fat, because kerosene was used as the heat-transfer medium in the calorimeter, and it dissolved the fat to form a thick jelly that could not be stirred readily. He also found that his data did not follow Siebel's rule between −40°C and +5°C.

The enthalpy of lean beef, some ocean fishes, venison, chicken, eggs, and breads over the range −40°C to +20°C has been measured with an adiabatic calorimeter (Riedel, 1956, 1957, 1959). By partially dehydrating the samples and measuring their enthalpy, he found that all free water was frozen at −40°C. Included in this work are data on the percentage of frozen water as a function of temperature and initial water content.

Other authors (ASRE, 1951, 1955; Tressler and Evers, 1957) have listed specific heat values for foods above and below freezing, but did not define the temperature range of these measurements.

The heat capacity of gelatin-water mixtures with gelatin concentrations of 9–100% over the interval between +25°C and −180°C was measured in an adiabatic calorimeter (Hampton and Mennie, 1932, 1934) previously used by other workers (Barnes and Maass, 1930). However, it is not possible to obtain accurate specific heat values from these data.

METHODS

The apparatus used for measuring the specific heat of samples at low temperatures is seen in Fig. 1. It consists of a rectangular block of polystyrene foam (9.5 × 10.2 × 25.5 cm) with a cylindrical hole (1.4 × 20.3 cm) in the center, into which an aluminum sample container is placed. A polystyrene plug is inserted at the top of the cell to prevent rapid heat transfer to the sample.

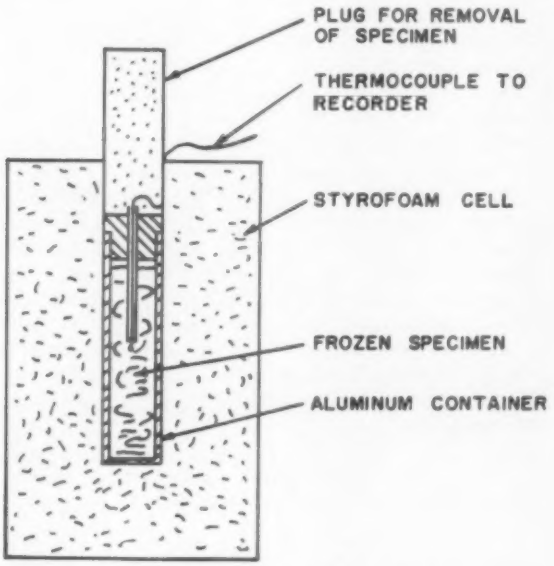

Fig. 1. Specific-heat apparatus.

The specimen container is filled with the material being measured (*ca.* 20 g), weighed, and then cooled rapidly (2–3 min) in liquid nitrogen. When temperature equilibrium has been reached in liquid nitrogen, the container is removed and quickly inserted into the polystyrene foam block maintained at room temperature. Temperature at the center of the specimen was measured continuously with a copper-constantan thermocouple and recorded over the desired range.

Calibration of the apparatus with a specimen of known specific heat is required, because it is necessary to know the rate of heat leak per unit area into the specimen during the slow warming period. For this purpose, a copper probe (ASRE, 1959) of known weight was used. By following the normal procedure of cooling in liquid nitrogen and recording temperature as a function of time during warming in the polystyrene cell, the heat leak through the polystyrene foam into the copper probe may be calculated.

The heat leak, Q, in cal/min at a particular temperature is equal to the product of the specific heat of the copper probe and its weight, multiplied by the rate of temperature change at that temperature. Once the heat leak has been determined over the temperature range of interest, the specific heat of any material of identical geometry may be determined, assuming that the rate of heat leak into the cell will be the same at any given temperature.

Once the rate of temperature change of the specimen container holding the material of unknown specific heat has been measured, it is possible to determine the specific heat of the specimen itself, using the value of Q previously obtained with the copper probe.

The specific heat of the entire specimen (Cps) at a given temperature is equal to the heat leak (Q) divided by the rate of temperature change ($\Delta T/\Delta t$) at that temperature and the weight of the entire specimen (Ws). Thus:

$$Cps = \frac{Q}{Ws\,\Delta T/\Delta t} \qquad (1)$$

The specific heat of the unknown material is determined by subtracting the heat capacity of the aluminum container (Cpa Wa) from the heat capacity of the unknown material plus container (Cps Ws) and dividing this value by the weight of the unknown material (Wx):

$$Cpx = \frac{Cps\ Ws - Cpa\ Wa}{Wx} \qquad (2)$$

By carrying out these computations for different temperatures, the change in specific heat with temperature is determined.

A specimen warming curve for ice is seen in Fig. 2. The rate of temperature change is determined by measuring the instantaneous slope of the warming curve at a point T, t.

RESULTS AND DISCUSSION

The reproducibility of measurements made with the technique described above is seen in Table 1, which shows the values for two measurements at a given gelatin concentration. The average error is 3.0% for a 26.4% gel, and 2.9% for a 39.1% gel. When the values for two determinations of pure gelatin are compared, the average error is larger than for aqueous gelatin solutions. This increase in error is probably due to the existence of a small thermal gradient throughout the sample. The gradient involved is the difference in temperature between the center of the specimen and the wall of the aluminum container. Air between the particles of the dry powder decreases the rate of heat transfer through the specimen, causing a temperature gradient. This gradient was determined by measurement to be about 5°C at −160°C, decreasing to less than 1°C at −30°C. In a gel, a continuous phase exists, and essentially no gradient is observed. Identical curves were obtained in repeated experiments.

The procedure outlined measures the total amount of heat absorbed by the system. Under conditions where an internal heat source does not cause a rise in temperature, such as the time during which the latent heat of fusion is being supplied, the values computed represent the total heat capacity of the system over the melting range. Under these conditions, the specific heat cannot be determined by experiment but can be estimated at higher temperatures by extrapolation of the linear portion of the curve to the intercept of the zero ordinate.

Table 1. Reproducibility of specific heat measurements.

Gelatin concentration (Wt–%)	Specific heat (cal/g)				
	−40°C	−80°C	−120°C	−160°C	% difference
26.4	0.441	0.375	0.319	0.218	
26.4	0.448	0.382	0.309	0.207	3.0
39.1	0.382	0.394	0.336	0.224	
39.1	0.385	0.403	0.328	0.213	2.9
100	0.312	0.260	0.209	0.158	
100	0.292	0.242	0.193	0.144	6.7

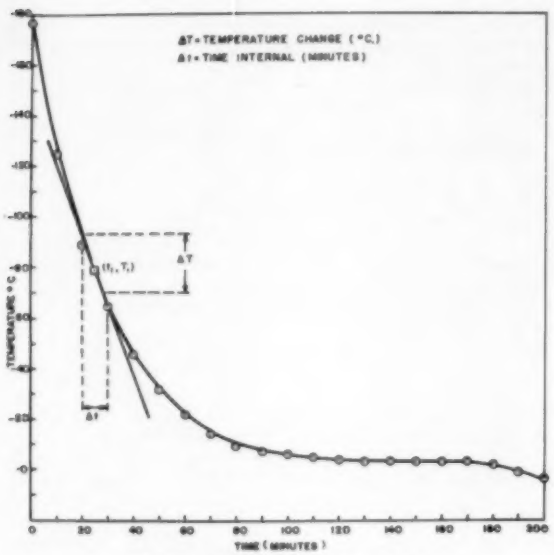

Fig. 2. Warming curve for ice.

The specific heat-temperature curve for ice as obtained by this method is almost identical to previously published curves (Dorsey, 1940), differing only by an average of 1.8%.

A typical specific heat curve for beef chuck is seen in Fig. 3. As the temperature increases, the specific heat increases. At about −50°C the heat capacity rises sharply. The extrapolation on the curve gives a measure of the specific heat of the sample between −50°C and its melting point.

With a method for measuring specific heat at low temperatures, it was possible to determine whether the specific heat of a complex system such as tissue was related to its composition. If so, the specific heat of a tissue could be predicted once the values of the specific heats of fat, protein, water, and salts, and the percentages of these constituents were known.

We selected gelatin as a representative protein and measured the specific heat at various aqueous dilutions. Also determined were the specific heats of some naturally occurring fats such as lard, beef suet, and butter (Table 2). A model system simulating the composition of beef was prepared by homogenizing 80 g of a 22% gelatin solution with 20 g of melted beef suet (containing 10% water) in a blender. This homogenate was then placed in the aluminum container and quickly frozen. The final composition of this system was 17.6% protein, 18% beef fat, and 64.4% water. Its specific heat was measured at low temperatures, and the values were compared with the specific heats obtained by computation from the components, that is, from the sums of the specific heat of each component multiplied by its fractional concentration (Table 3).

Table 2. Specific heat values of ice, fats, protein (gelatin), used to calculate the specific heats of foodstuffs.

Sample	−40°C	−80°C	−120°C	−160°C
Ice[a]	0.438	0.368	0.298	0.232
Gelatin[a]	0.310	0.250	0.201	0.151
Beef fat[a]	0.550	0.371	0.239	0.202
Lard[a]	0.383	0.290	0.245	0.211
Butter[a]	0.465	0.322	0.264	0.240
Sodium chloride[b]	0.194	0.185	0.171	0.149
Dextrose[b]	0.230	0.184	0.139	0.112

[a] Values obtained by experiment.
[b] These values were obtained from the International Critical Tables.

Table 3. The specific heat of a model system[a] as obtained by direct measurement compared to the specific heat calculated by constituent analysis.

	Cp		
°C	Computed[b]	Observed	Observed/computed
−40	0.435	0.468	1.07
−80	0.348	0.387	1.11
−120	0.271	0.307	1.13
−160	0.212	0.228	1.13
			Av. 1.11 ± 0.03

[a] Composition of the model system was 64.4% water, 17.6% protein, and 18.0% fat.
[b] The sum obtained after multiplying the specific heat of each component by the weight fraction present.

The computed values are consistently lower than the observed values by a factor of 1.11 ± 0.03. A similar difference, a factor of 1.14 ± 0.06, was observed when meats of natural occurrence, in contrast to the model system just described, were tested by comparing measured specific heat values with computed values (Table 4). These meats differed considerably in water, fat, and protein content. The data indicate that it is possible to predict the specific heat values of meats using the correction factor (1.14) with an average error of 4.4% and a maximum error of 10.5%. Had the estimate of the specific heat been based on the water amount only, the error would have been about 35%.

The correction factor for the computed specific heats of the two fish products measured is 1.09 ± 0.05, a value quite similar to that obtained with both meats

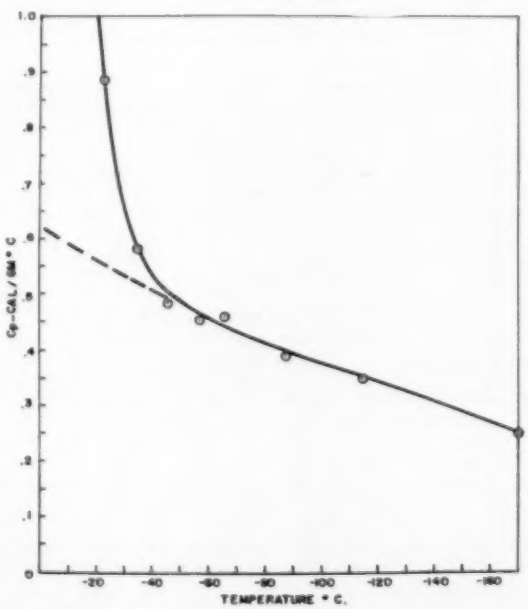

Fig. 3. The specific heat of beef chuck.

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Table 4. Comparison between measured and computed specific heats for several different meats.

	Observed Cp/computed Cp[a]				
°C	Beef liver	Beef round	Beef loin	Beef chuck	Smoked cooked ham
−40	1.15	1.14	1.11	1.18	1.14
−80	1.08	1.09	1.18	1.09	1.23
−120	1.09	1.10	1.24	1.11	1.26
−160	1.18	1.05	1.18	1.05	1.19
				Av. value	1.14 ± 0.06

[a] The sum obtained after multiplying the specific heat of each component by the weight fraction present.

and the model system (Table 5). Here, the average error, using the correction factor of 1.09, is 2.7%.

The difference between the calculated and observed values could be due to vitreous ice in the frozen system. In a gelatin gel of the same concentration of protein as found in the meats and fish tested, most of the free water has formed normal hexagonal ice at these low temperatures, but a small yet significant amount of this water is present as vitreous ice (Dowell *et al.*, 1961). In general, the specific heat of a vitreous solid is higher than that of the corresponding crystalline solid (Partington, 1952). If the assumption is made that the specific heat of vitreous ice is higher than the specific heat of crystalline ice, we would anticipate that the computed specific heat values for the model system or meats would be lower than the observed values. Since this factor was not considered in our computations, the difference in the specific heat of vitreous and crystalline ice might account for the discrepancy in the two values.

When the differences between the computed and the observed values were measured for the specific heats of the two dairy products, cheddar cheese and ice cream, the computed values were found to be higher than the observed values (Table 5). The ratio of the observed to computed specific heat is 0.87 ± 0.06 for cheddar cheese and close to unity, 0.96 ± 0.11, for ice cream.

In ice cream, which has a low protein content (4%), very little or no vitreous ice would be formed. Therefore, the computed and observed specific heats should be essentially the same. The value of 0.96 obtained for the ratio of the observed to the computed specific heat would support this hypothesis. Cheddar cheese, on the other hand, contains essentially equal amounts of water and protein, and a high percentage of the water present would be bound to the protein.

Using the values for the specific heat of bound water between 0 and −70°C (Hampton and Mennie, 1932) and our specific heat values for ice, the ratio of the specific heat of bound water to ice at −40°C is 0.92, a value almost identical with that obtained for the ratio of the observed to the computed specific heat in cheddar cheese. As the temperature is lowered, the specific heat of the bound water decreases more rapidly than the specific heat of ice. This decrease may account for the diminishing ratio of the observed to computed specific heats for cheddar cheese.

Table 5. Comparison between measured and computed specific heats for some fish and dairy products.

	Observed Cp/computed Cp[a]			
°C	Raw green shrimp	Cod fish cakes[b]	Ice cream	Cheddar cheese
−40	1.09	1.11	0.83	0.92
−80	1.09	1.09	0.92	0.90
−120	1.07	1.06	1.01	0.86
−160	1.02	0.97	1.09	0.79
Av. values	1.09 ± 0.05		0.96 ± 0.11	0.87 ± 0.06

[a] The sum obtained after multiplying the specific heat of each component by the weight fraction present.

[b] The computed specific heat values for this product were obtained by measurement of the ether extract (7.7%), the ether nonextractables (26.8%), and water (65.5%).

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Relationship of Post-Mortem Acidity and Temperature[a]

(Manuscript received June 10, 1960)

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SUMMARY

Various combinations of temperature and acidity were studied in relation to change in muscle structure. Soft, exudative tissue resulted when pH was decreased to about 5.4 (lactic acid, *ca.* 1%) while tissue temperature remained above 25°C. Chilling the tissues rapidly prevented the extreme pH variations and muscle structure alterations. Rapidly chilling carcasses through submersion did not give the same degree of improvement, but substantially retarded pH decrease immediately post-mortem. The result was a material improvement of color intensity and water-binding capacity in fresh tissue as well as reduced gelatin formation in the canned product.

THE NORMAL RATE of post-mortem anaerobic glycolysis usually permits marked reduction in muscle temperature before substantial acidity develops (Bate-Smith, 1948; Bendall, 1951). Extreme variations, however, have been noted in the rate of decrease in pork muscle pH during the immediate post-mortem period (Briskey and Wismer-Pedersen, 1961; Briskey *et al.*, 1959, 1960; Ludvigsen, 1954; Wismer-Pedersen, 1959). More recently Wismer-Pedersen and Briskey (1961) reported several post-mortem pH patterns and suggested that the effects of rate of glycolysis on muscle structure were influenced greatly by the concomitant temperature of the muscle.

This paper reports: 1) the temperature and acidity combinations that influence muscle color, structure, and pH; and 2) preliminary attempts to alter the pH pattern through a change in rate of chilling.

EXPERIMENTAL

Experiment I. *Alterations in muscle structure at different temperatures.* Two carcasses of low pH_1 and two carcasses of high pH_1 were obtained from the Roskilde Bacon Factory immediately after evisceration (Wismer-Pedersen, 1959). The *longissimus dorsi* muscles from the right sides (10th rib to last lumbar vertebra) were removed, divided into three equal sections, and labeled 2A, 2B, and 2C. The radiometer pH meter (model 31) was used in this experiment to obtain continuous recordings of pH and temperature as described by Wismer-Pedersen and Briskey (1961). The pH electrodes and thermocouples were uniformly placed in the medium of each section. The sections were placed in polyethylene bags and subjected to the following treatments:

1) Section A was placed for 1 hr in a thermostatically controlled Plexiglas-styrofoam chamber at 37°C. The sample was removed from the chamber and held 24 hr at 3°C.

2) Section B was placed in an ice-salt mixture at −2°C. When the muscle temperature decreased to 3°C the sample was removed from the chamber and held 24 hr at 3°C.

3) Section C was held 24 hr in a 3°C atmosphere to simulate a normal rate of chilling.

Electrodes and thermocouples were also inserted in the *longissimus dorsi*, of the left side, at the area of the 12th rib to serve as a dual control.

Samples were removed at 1, 2, 4, 6, 10, and 24 hr, and loose-water numbers were determined at each period according to the method of Wismer-Pedersen (1959). The samples were then immediately frozen in dry ice and held for subsequent analysis of lactic acid according to a slight modification of the method outlined by Barker and Summerson (1941). Sections 2A, 2B, and 2C were given the same treatment indicated above, and used to estimate color change and "drip" loss. The change in the intensity of the color was recorded with the Elrepho Reflectance Meter at 540 mμ. A magnesium-oxide standard was used, with 100 as pure white.

Experiment II. *Further studies on the combined effect of acid and temperature on meat color and pH.* A) *Fresh muscle mince with three different solutions. Psoas major* muscles with high pH_1 were excised within 20 min of being bled, and passed through a household meat grinder (2-mm holes). The mince was divided into three parts, and each portion was thoroughly mixed with one of the following solutions at the concentration of 5 ml per 50 g of mince: A, distilled water; B, lactic acid, 1%; and C, 0.005*N* iodoacetate. Portions of the minced product (25 g) were transferred to Petri dishes, placed in Cryovac bags, and subsequently submitted to vacuum. Samples from each treatment were: a) chilled 1 hr in an ice-salt mixture at 0° to −2°C, b) retained 1 hr at 37°C, and c) held 1 hr under normal cooler conditions (3°C). Control samples for each mixture and treatment were subsequently held 24 hr at 3°C. Color intensity and pH were recorded before and after treatment by the methods described in Experiment I.

B) *Fresh muscle mince with various lactic acid concentrations.* The mince was prepared by passing the tissue through a household meat grinder (2-mm holes). The mince was immediately mixed with 5-ml quantities of solutions containing the following lactic acid concentrations: A, 1.25%; B, 1.00%; C, 0.75%; D, 0.50%; E, 0.25%; and F, 0.005*N* iodoacetate. The samples from each mixture were transferred to Petri dishes, placed in Cryovac bags, subjected to vacuum, and immediately placed in temperature-control chambers, where they were held 1 hr at the following temperatures: 20, 25, 30, 35, and 40°C. The samples were then removed and held 24 hr at 3°C. Changes in color and pH were recorded by the methods described in Experiment I.

C) *Chilled muscle sections with various ultimate pH values.* Used in this study were chilled *psoas major* muscles of varying pH (5.35, 5.65, 5.90, 6.20). Thin sections (⅜ in.) were removed and placed in small Petri dishes. These units were placed in Cryovac bags, submitted to vacuum, and immediately placed in temperature-control chambers, where they were held 1 hr at the following temperatures: 20, 25, 30, 35, and 40°C. Color intensity and pH before and after treatment were recorded by the methods described in Experiment I.

Experiment III. *Effect of rate of chilling on fresh and processed products.* Used in this study were 6 Danish Landrace pigs ranging in weight from 85 to 95 kg. The pigs were taken to the Roskilde Bacon Factory, where they were electrically stunned and slaughtered. Within 30 min of bleeding, the left sides were weighed and resistance thermometers were inserted in the medium of the *semimembranosus* (ham), *longissimus dorsi* (loin), *pectoralis profundus* (belly), and *serratus ventralis* (shoulder). The sides were then placed in polyethylene bags and the air was removed with a household vacuum apparatus. The prepared sides were immediately submerged in a −2°C brine. This cooling process was continued until the ham

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center achieved a temperature below 20°C. The sides were immediately removed and placed in a chilling room (Danish Meat Research Institute) with an air temperature of +3°C.

The right sides, as controls, were placed in the 3°C atmosphere within 45 min of bleeding. Continuous recordings of pH and temperature were facilitated with the specialized pH meter (Radiometer model 31).

After the carcasses were chilled 24 hr at 3°C, they were removed from the chilling room and weighed. Chilled samples were later removed from the *longissimus dorsi* at the second lumbar region and held for immediate analysis of lactic acid concentration (Barker and Summerson, 1941), color intensity (Briskey and Wismer-Pedersen, 1961) and water-holding capacity (Wismer-Pedersen, 1959).

Processing treatment. The carcasses were fabricated into hams, shoulders, and loins as recommended by the 5th Reciprocal Meats Conference Committee (1952). These cuts were ultimately stitch-pumped with a normal curing brine (salt, nitrate, nitrite, and sodium tripolyphosphate) to 8% of their weight. After curing for three days, the cuts were boned, trimmed, and canned (66°C). Sausage emulsion was prepared from the tenderloin and belly. The emulsion was canned in cylindrical cans and sterilized 1 hr at 108°C. The cans were stored one month at 4°C, and ultimately examined for gelatin content by the method of Wismer-Pedersen (1959), color intensity as suggested by Briskey and Wismer-Pedersen (1961), organoleptic quality as reported by Wismer-Pedersen (1960), salt development by a modification of A.O.A.C. (1955) method, and nitrose-haem pigment concentration as outlined by Hornsey (1956).

RESULTS

Experiment I. *Acidity-temperature-time relationships in regard to alterations in muscle structure during post-mortem chilling.* Fig. 1 shows chilling rates and pH patterns for the loin sections from carcasses with high pH_1. The rapid chilling of B sections produced pH patterns characteristic of the type 2 muscles, described by Wismer-Pedersen and Briskey (1961). The B sections showed "drip" loss values of about 1.37%, and color intensity values of 16.15 reflectance units, and were adjudged normal in structure (Briskey *et al.*, 1960). The retention of heat for one hour in the A sections, however, produced pH patterns characteristic of the type 4 muscles (Wismer-Pedersen and Briskey, 1961). The A sections showed "drip" loss values of about 4.60%, and color intensity of about 28.0 reflectance units, and were adjudged pale, two-toned in color, and soft in structure (Briskey *et al.*, 1960). The C sections (chilled in a 3°C cooler) were intermediate in color and "drip" loss (21.00 reflectance Units, 3.66% loss). Because of the "drip" loss the LW (loose-water) values were extremely variable.

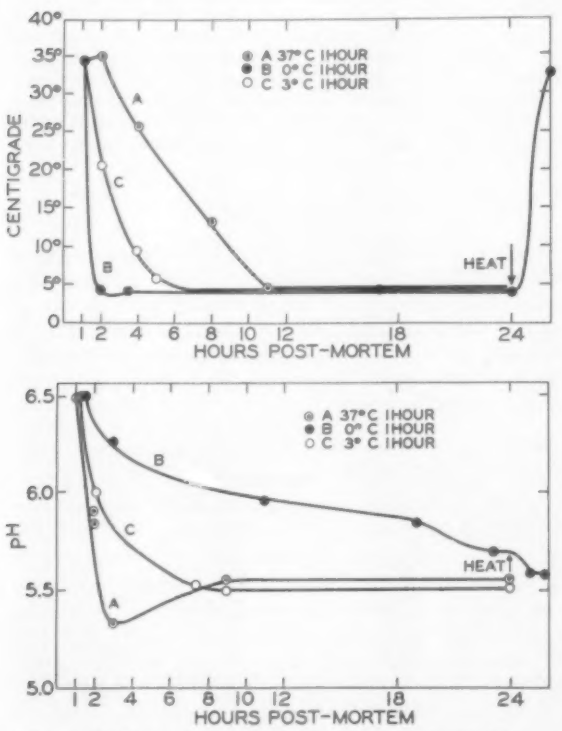

Fig. 1. Chilling rates and pH patterns for high-pH_1 loins.

Fig. 2 shows chilling rates and pH patterns for the loin sections from carcasses with low pH_1. The rapid chilling of section B prevented continuation of the severe drop and the elevation in pH that Wismer-Pedersen and Briskey (1961) noted to be characteristic of the type 4 muscles. These sections had "drip" loss values of 2.1%, and color intensity values of 21.7 reflectance units, and were adjudged normal in structure (Briskey *et al.*, 1960). The retention of heat for 1 hr in sections A of these muscles, however, exaggerated the severity of the pH depression and elevation reported for the type 4 muscles (Wismer-Pedersen and Briskey, 1961). These sections lost about 6.0% drip and decreased in color intensity to 28.0 reflectance units. Consequently, these sections were adjudged abnormally pale and inferior in structure. The C sections, though improved in comparison to the A sections, were still inferior in quality, and showed color intensity values of 24–25 reflectance units. These sections were adjudged pale, two-toned in color, and soft in structure.

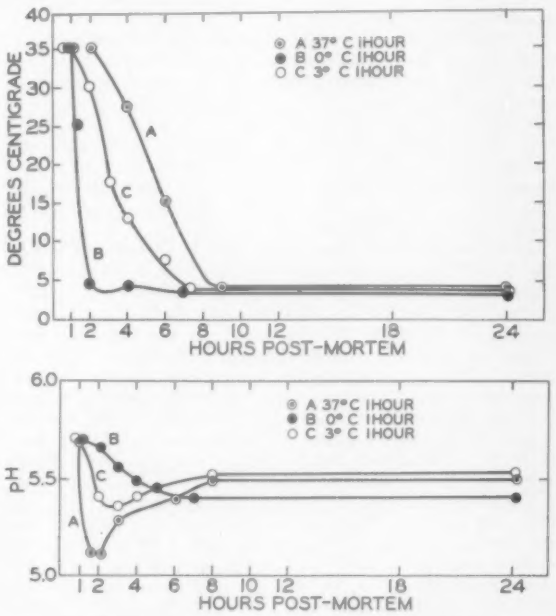

Fig. 2. Chilling rates and pH patterns for low-pH_1 loins.

Although the increases in lactic acid due to heat retention were not as pronounced as those that occurred normally at elevated temperatures in previous experiments, lactic acid formed significantly faster in the A sections than in the B and C sections during the initial treatment. This increase, however, was of the greatest magnitude in the A sections from the low-pH_1 carcasses.

Experiment II. *Further studies on the combined effect of acid and temperature on meat color and pH.* A) *Fresh muscle mince with three different solutions.* Fig. 3 shows the effects of solution and temperature on the color intensity of fresh muscle mince. Group A (water) showed the usual effects of

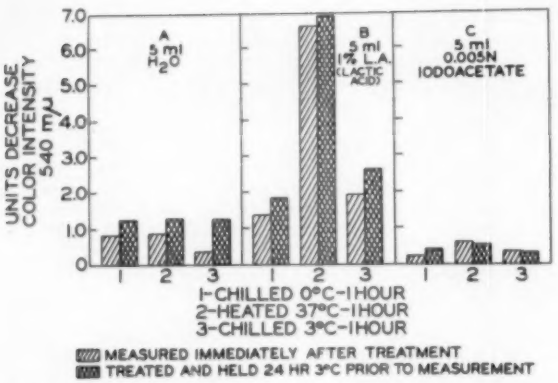
UNITS DECREASE COLOR INTENSITY 540 mμ
A 5 ml H2O
B 5 ml 1% L.A. (LACTIC ACID)
C 5 ml 0.005N IODOACETATE
1-CHILLED 0°C-1HOUR
2-HEATED 37°C-1HOUR
3-CHILLED 3°C-1HOUR
MEASURED IMMEDIATELY AFTER TREATMENT
TREATED AND HELD 24 HR 3°C PRIOR TO MEASUREMENT

Fig. 3. Combined effects of acidity and temperature on muscle color.

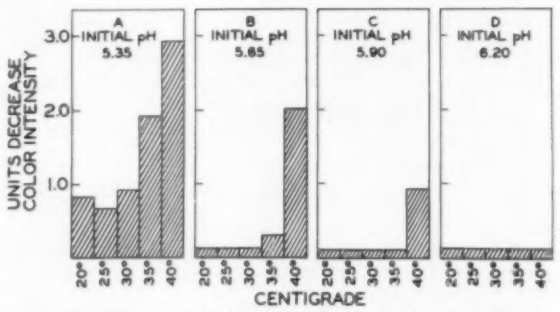

Fig. 4. Influence of pH on the sensitivity of muscle color at various temperatures.

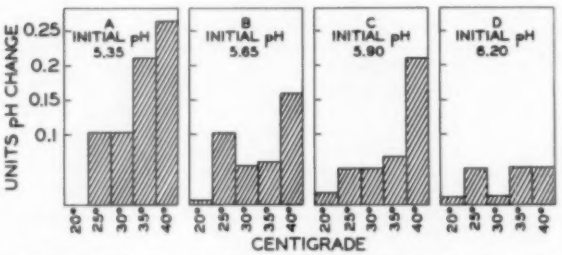

Fig. 5. Influence of pH on the sensitivity to change in pH at various muscle temperatures.

acid formation and chilling time on muscle color. The samples in Group B (lactic acid) showed a greater change in color intensity at all temperatures, with the most pronounced change appearing in the mince held 1 hr at 37°C. The iodoacetate solution (Group C) blocked acid formation and contributed to retention of the intense color of the fresh muscle mince.

B) *Fresh muscle mince with various lactic acid concentrations.* In this experiment, the muscle preparations with at least 0.75% added lactic acid showed the greatest sensitivity to temperatures above 25°C. This sizable reduction in color intensity was especially apparent in samples with 1.25–0.75% added lactic acid when heated to 30°C. Conversely, the preparations that received 0.25% or 0.50% lactic acid showed only minor reductions in color intensity until they were heated above 35°C. The samples that received 0.005*N* iodoacetate resisted the change in color regardless of heat treatment.

C) *Chilled muscle sections with various ultimate pH values.* The data in Figs. 4 and 5 distinctively imply a greater sensitivity to heat in the low-pH muscle. Section A (pH 5.35) showed minor changes in color intensity at 20, 25, and 30°C, and sizable reductions at 35°C. There were no color intensity changes in section B (pH 5.65) until the samples were heated to 40°C. With an ultimate pH value of 5.90 (Section C) there were only minor reductions in color intensity at 40°C, whereas the samples in section D (pH 6.20) resisted changes throughout the temperature treatments. It can also be inferred from the data in Fig. 5 that the pH changes show reasonable parallelism to the reductions in color intensity.

Experiment III. *Effect of rate of chilling on fresh and processed products.* Fig. 6 shows representative chilling rates for various muscles in the treated and control sides. The chilling rates in the ham centers were only moderately accelerated by submersion in the cold brine. The ham surface, loin and belly, however, were chilled much faster in these treated sides. Table 1 summarizes the effect of chilling rate on pH pattern, lactic acid formation, loose-water development, and color retention. These data imply that there were no substantial differences in ultimate (24-hr) lactic acid concentrations or pH values. The loose-water values were significantly decreased in the submerged sides, and the color intensity values were significantly increased.

There were no significant differences in salt content, nitrose-haem pigment formation or gelatin percentage of the canned products. Definite trends toward decreased salt and gelatin in the treated products were, however, quite apparent. The sausage emulsions from the submerged sides contained about 4% less gelatin than their respective controls. Organoleptic evaluations showed no significant differences in saltiness, taste, and texture as a result of treatment. The color index values in the canned hams and loins tended to be lower (darker) from the submerged sides.

DISCUSSION

It is particularly noteworthy that it appears possible to produce pale, exudative tissue by retaining body temperature for an extended period. It is equally pertinent to point out that the rapid chilling of sections with low pH_1 values prevented continuation of the severe depression and elevation in pH

Table 1. Color intensity and water-holding capacity of fresh *longissimus dorsi* in relation to pH and chilling rate.

	Control sides						Submerged sides					
Carcass no.	pH (30-min)	pH (24-hr)	pH pattern[a]	LW[b] (mg/g)	Lactic acid (%)	Color index[c]	pH (30-min)	pH (24-hr)	pH pattern[a]	LW[b] (mg/g)	Lactic acid (%)	Color index[c]
1	5.90	5.40	4	507	1.13	22.8	5.90	5.50	3	456	1.07	18.2
2	6.35	5.45	4	475	1.09	19.9	6.35	5.65	2	459	1.06	18.3
3	6.45	5.60	4	490	1.15	20.3	6.45	5.60	3	436	1.15	16.7
4	6.45	5.55	4	479	1.04	23.0	6.45	5.60	2	431	1.08	17.8
5	5.55	5.58	4	506	1.18	32.8	5.55	5.48	3	442	1.11	26.5
6	6.45	5.42	3	461	1.10	19.5	6.45	5.65	2	383	1.04	17.4
Mean		5.50		486	1.11	23.1		5.58		435**	1.08	19.2**

[a] pH patterns (Wismer-Pedersen and Briskey, 1961): 1, extremely slow decrease; 2, slow gradual decrease; 3, relatively rapid decrease; 4, sharp, significant, immediate decrease.
[b] Loose-water (mg/g tissue) Wismer-Pedersen (1959).
[c] Color index (Zeiss, Elrepho Lichtelektrisches Remissions photometer).
** Significantly different from the control sides at the 1% level.

(Briskey and Wismer-Pedersen, 1961), and thus the sections were normal in structure. This finding is in line with the inference (Wismer-Pedersen, 1959) that muscle temperature and pH combinations may markedly influence its ultimate appearance, and also opens a new area for research on the relationship of acidity-temperature-time and muscle structure.

Although various carcass chilling rates were obtained, it was nevertheless obvious that it was difficult to apply this principle of rapid chilling in muscle sections to the intact carcass (Am. Soc. Refr. Engrs., 1959; Callow, 1938; Jul, 1959). It was also difficult, under normal conditions, to secure carcasses from the slaughter line before the initial impact of the acidity-temperature-time factor in those carcasses inclined to possess an extremely rapid rate of anaerobic glycolysis and consequently to follow drastic pH patterns

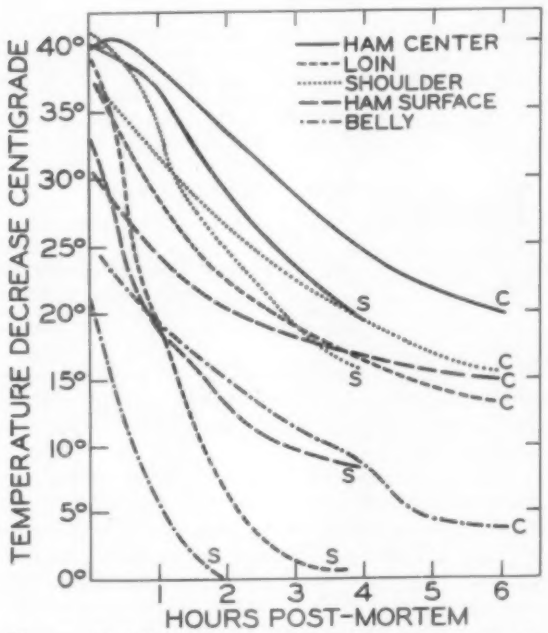

Fig. 6. Variations in chilling rates of control and submerged sides.

(depression and elevation) (Briskey and Wismer-Pedersen, 1961). In spite of these problems, it appears valid to study the principle in excised muscle sections (Fremery and Pool, 1960).

The data in Figs. 3 and 4 support the work of Bate-Smith and Bendall (1945) and Bendall (1946, 1947), who found pH elevations to be greatest in low-pH solutions. In the present experiment, temperatures to 40°C were not critical if pH values were above 6.2. Temperatures above 35°C appeared to be influencing factors if the muscle tissue possessed a pH value below 5.90. This suggests that if the pH_1 decreases below 5.90 the muscle will undoubtedly be altered in structure. Similarly, a temperature above 30°C is critical if the pH value is below 5.6. The data are of comparable importance in interpreting muscle changes in intact carcasses that, in effect, reveal various combinations of acidity-temperature during post-mortem chilling. Although the procedures used were not successful in appreciably accelerating the chilling rate in the intact carcass, the improvements in chilling rate were accompanied by concomitant improvements in pH change, water-binding capacity, and color development. These improvements were observed in the *longissimus dorsi* muscles, which, because of the thickness and exposure factors, were chilled at an accelerated rate. The accelerated chilling rate had no significant effect on total lactic acid content, but it did reduce the rate of lactic acid formation.

The "open" and "closed" structures as defined by Callow were specifically demonstrated in these experiments. The rapidly chilled sections and the submerged sides reflected the "closed" structures, which were evidenced in the sections by higher pH_2, darker color, and improved water-binding capacity.

Although no practical aspects were implied, these observations are of pertinence since pork muscle frequently exhibits considerable acidity at high temperatures (37–40°C) and subsequently reflects the pale, watery condition. When the chilling rate was substantially altered, as in Experiment I, the adverse change in structure was significantly reduced. Comparable work with intact carcasses showed that slight alterations in chilling rate prevented sharp reductions and elevations in pH, and therefore reflected an improved fresh muscle quality. The rate of heat removal from 40 to 20°C was of great importance in determining the effect of lactic acid formation on water-binding capacity and gelatin formation.

Further studies must be conducted to determine the specific effects of these temperature-acidity combinations on the nature of the individual muscle proteins.

Acknowledgment

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Stabilized Raisins for Dry Cereal Products

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(Manuscript received September 12, 1960)

SUMMARY

Materials for stabilizing raisins against loss of moisture were studied. Beeswax was the best moisture barrier of the edible coatings tested. Diluting beeswax with cottonseed oil or other vegetable oils increases its permeability to water vapor. A method for the application of a moisture barrier to raisins was devised. The raisins are dusted with starch and then coated with beeswax or a mixture of beeswax and vegetable oil. Untreated raisins stored with bran flakes became hard and inedible in about one-fourth of the time required for wax-coated raisins. Texture measurements with a shear press indicate increasing toughness as raisins are dried out.

THE SHELF LIFE of prepared breakfast-food flakes containing raisins is considerably shortened by transfer of moisture from fruit to cereal. Eventually, the flakes lose crispness and the raisins become hard and less palatable.

Attempts to extend the shelf life of the raisin-cereal product have had but moderate success. Some manufacturers have reduced the vapor pressure differential between the two constituents by increasing the moisture content of the bran flakes. Coatings of beeswax, raisin oil, and sugar applied commercially have not been effective moisture barriers. Breaks and pin holes are seen in the coating under a low-power microscope. However, a thin film of beeswax applied in the manner described below is an effective moisture barrier capable of extending shelf life by a factor of three or more.

Raisins as received from the processor normally have small droplets of moisture or syrup on the surface. These are particularly apparent at points where the skin is broken. Mechanical or insect damage to the grapes also results in tiny droplets of liquid on the outside of the raisins. When a waxy coating is applied, these droplets are not covered; thus, a small hole is left in the wax layer and moisture can escape from the interior of the raisin.

Two approaches to stabilizing raisins for dry cereal were considered: adding edible humectants, and coating the surface with a moisture-vapor barrier. Preliminary tests showed humectants to be unsatisfactory, either because of changes in flavor or stickiness of the treated raisins. A moisture-impervious film, in contrast, seemed to offer considerable promise. Accordingly, a number of materials were screened. Dusting raisins with starch and then dipping them in molten beeswax proved effective. The process has been carried through pilot-plant development. Details of pilot-scale operations will be described later.

EXPERIMENTAL

Materials. Test fruit was regular Thompson Seedless raisins containing 14–18% moisture. Commercial bran flakes containing 3.2% moisture were stored in a low-humidity room until

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used. Commercial edible corn, rice and potato starches, pectin, proteins and modified proteins, acetylated glycerides, animal and vegetable fats and waxes, and natural and synthetic emulsifiers were tested. The beeswax used was pure edible beeswax of technical or food grade. A simple cobaltous chloride hygrometer (Brekke and Watters, 1958) facilitated rapid testing of many materials for their resistance to passage of moisture vapor. A thin film of the material tested separates a strip of paper impregnated with $CoCl_2$ and a vial of water. The number of hours required for the $CoCl_2$ strip to change from blue to pink is recorded. These values are interpreted as inverse indices of the permeability of the materials to water.

Methods. Many of the non-lipid materials were applied as solutions and allowed to dry on the surface of the raisins as a film. Lipid materials were applied in liquid form; those that are solid at room temperature were applied in a bath at elevated temperature. Starch was applied dry by merely tumbling the raisins and starch together in a container. Satisfactory materials for precoating raisins are corn, potato, and rice starches. Dusting raisins with dry starch conditions the surface so that a continuous layer of lipid material can be applied. The excess of starch involves the water in the surface droplets in such a way that a dry surface develops which is compatible with lipids. Beeswax was applied after starch precoating by immersing raisins for 2–3 min in a bath of molten beeswax (210–220°F). The raisins were thus warmed to near the temperature of the bath, and excess beeswax was easily removed by tumbling the raisins on a screen or on an absorbent surface such as a paper towel. The laboratory-scale tests were run on 2–3 lb of raisins at the time.

RESULTS AND DISCUSSION

Materials such as proteins, pectin, liquid oils and waxes, and natural and synthetic emulsifiers were ineffective as moisture barriers. As can be seen in Table 1, a beeswax film is manyfold more resistant to the passage of moisture than the above materials.

Table 1. Time required for moisture vapor to penetrate films of test materials in cobalt chloride hygrometer.

Test material	Hours
Filter paper	2
Pectin	2
Casein	5
Cottonseed oil	18
Coconut oil	19
Cetyl alcohol	21
Jojoba oil	24
Lecithin	55
Acetostearin	55
Beeswax	473

The most effective moisture barrier was a film of beeswax applied to raisins pretreated with dry starch. Less effective was beeswax diluted with liquid oils such as cottonseed oil or raisin oil. The liquid oils apparently diluted the effect of the solid beeswax.

Treated raisins and untreated controls were stored at 0% and 20% relative humidity. Changes in weight indicate loss of moisture. As can be seen in Fig. 1, the beeswax film provided the greatest protection against moisture transfer. Similar results were reported by Schultz *et al.* (1949).

Raisins and bran flakes (3.2% moisture) were stored at 70°F in hermetically sealed glass containers. Fig. 2 shows the results. The uncoated raisins in 5 weeks were down to 11% moisture, considered by many in the cereal-products business to be the acceptable minimum. At 5 weeks the beeswax-treated raisins

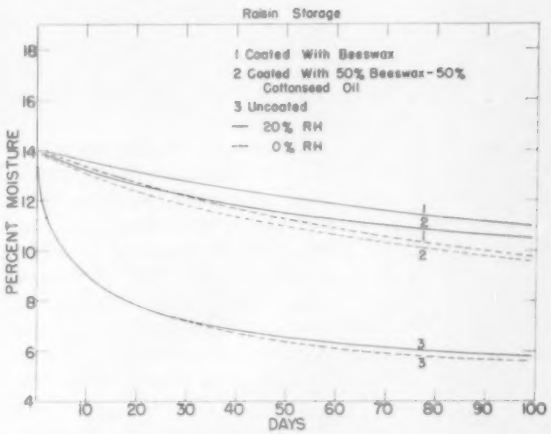

Fig. 1. Moisture content of raisins held at 0% and 20% relative humidity.

still contained 14.5–16% moisture. At 90 days, raisins coated with 100% beeswax were at higher moisture levels than those coated with 50% beeswax.

Raisin bran flakes were "manufactured" by addition of raisins 1:3 (w/w) to commercial bran flakes. The mixtures were then returned to retail packages, resealed, and placed at constant temperature (70°F). Relative humidity varied between 25% and 75% (av about 50%) during storage. The coated raisins lost moisture considerably more slowly than the uncoated (Fig. 3), and pure beeswax was more effective than 1:1 beeswax and cottonseed oil. The moisture content of the untreated raisins decreased from 17.5% to 11% in 2 weeks; those coated with a 100% beeswax film had an initial moisture content of 16.5%, and 9 weeks passed before they had dried to 11% moisture. In this experiment, packaging material undoubtedly plays a role in the moisture transfer phenomenon, and atmospheric conditions likewise could exert an effect rather too variable for easy analysis.

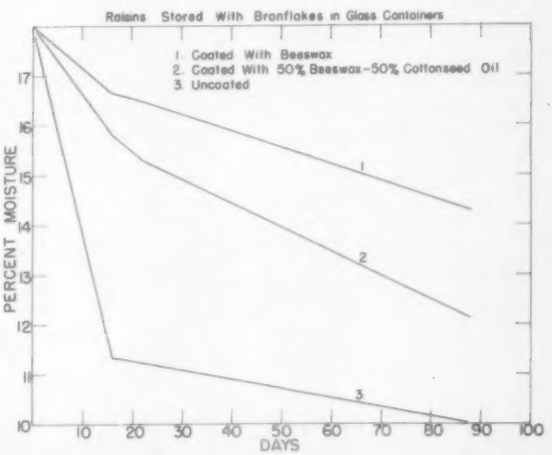

Fig. 2. Moisture content of raisins stored with bran flakes in glass containers.

A modified shear press was used to measure and compare the texture of regular and "midget" raisins at various moisture contents (Kramer and Backinger, 1959). "Midgets" are small-size raisins frequently used in cereal products and baked goods. The results are in Fig. 4. Raisins from the raisin bran flake experiment were tested on the shear press at 9 weeks. The results are expressed as pounds of force necessary to shear through a sample of raisins: untreated (9.5% moisture), 1755; 50% beeswax–50% cottonseed oil (10.0% moisture), 955; 100% beeswax (11.0% moisture), 1430. The raisins coated with beeswax and 1:1 beeswax and cottonseed oil appeared softer and more edible than the controls. The low results for the raisins coated with beeswax and vegetable oil may be due to the effect of the liquid component on the shearing action of the press.

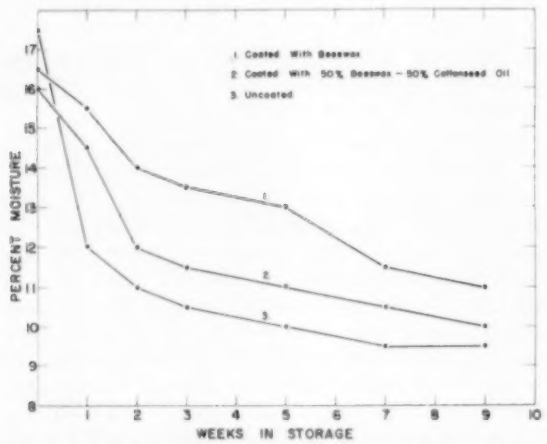

Fig. 3. Moisture content of raisins from "raisin bran flake" product packed in cardboard box with waxed-paper inner liner.

The thickness of the coatings applied by mechanical applicators can be controlled by the conditions maintained in the equipment. Thin coatings (only 1.2% by weight of the raisin) have been applied. According to (Lovegren and Feuge, 1954; Landmann *et al.*, 1960) the amount of moisture vapor transferred through a film of lipid material was affected only slightly by moderate changes in film thickness; our results generally confirm this. Coatings of about 2% of the weight of the raisin can be expected to give results as reported in this paper.

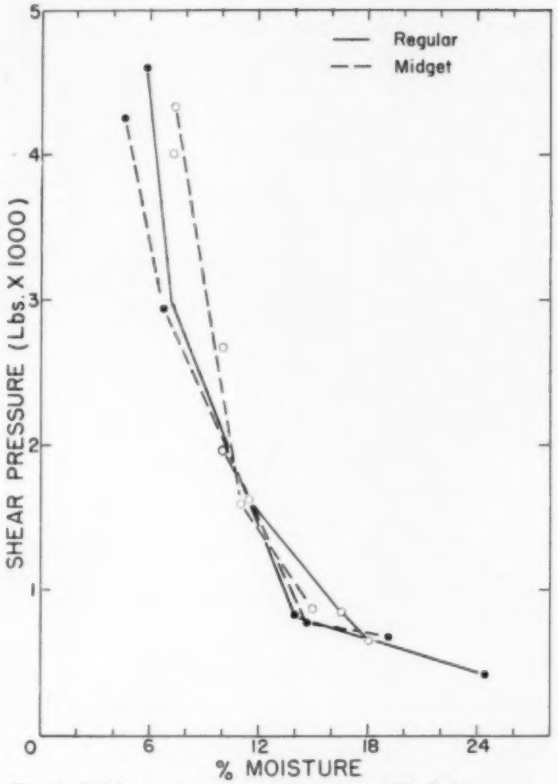

Fig. 4. Raisin texture measurements by modified shear press.

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Radiation Preservation of Milk and Milk Products. XII.

Radiation Sterilization of Milk in a Rotary-Disc Apparatus[a]

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(Manuscript received October 14, 1960)

SUMMARY

An apparatus was constructed that allows the production of sterile concentrated (3:1) milk that is essentially free of off-flavor. Polyphosphate-treated milk processed in this manner may be stored at 30°C in sterile form without gelation for at least 6 months.

It is possible that the apparatus may find practical use in the electron irradiation of other liquids of suitable viscosity.

DEVELOPMENT of a process for radiation sterilization of milk has been a subject of research in this laboratory for several years. The principal problems encountered, aside from construction of equipment that would ensure a sterile product, were the changes in milk brought about by irradiation. Some of these changes were the production of off-flavors, and the development of gelation and browning in storage.

In an effort to overcome the production of off-flavors, a radiation distillation apparatus was developed (Wertheim *et al.*, 1959) that allowed removal of the off-flavors. The resulting product, though free of off-flavor, was of limited value since it could not be obtained in a sterile condition.

With the apparatus to be described, however, concentrated (3:1) milk has been produced in sterile form essentially free of off-flavor. It is possible that this same apparatus may have application in electron irradiation of other liquid materials of suitable viscosity.

Factors considered in the design of the apparatus were the desirability of continuous operation, ease of cleaning and sterilization, prevention of corrosion, penetration of the electron beam, and removal of off-flavor. The energy level of electrons produced from the General Electric accelerator available in the Department of Food Technology was 0.8 Mev. This necessitated irradiation of a thin film (about 2 mm thick) of milk in order to obtain a sterilization dose.

A tubular vessel for irradiation of liquids and gases under pressure was described by Trageser (1958). Since this apparatus required electron energies higher than 1 Mev, and since its use with milk might lead to problems in obtaining even flow as well as to fouling of its interior by milk, its adaptation to the current problem was not considered feasible.

The apparatus constructed, which has produced sterile concentrated milk essentially free of off-flavor, consists of three basic parts: 1) an irradiation chamber; 2) an evaporator in which off-flavors are removed by aseptic vacuum distillation and the milk is diluted to the desired concentration; and 3) an aseptic sampler. The irradiation chamber contains a rotating disc, or turntable, onto which is continuously fed a thin layer of concentrated (4:1) milk. The layer of milk is moved by the turntable through the electron beam at a rate that allows its sterilization, and drained into the evaporator. As soon as the evaporator is filled with irradiated milk it is set up for distillation. During distillation the volume of liquid in the evaporator is maintained by addition of sterile water. Upon completion of distillation the resulting sterile irradiated milk is drained from the evaporator into appropriate containers in the aseptic sampling chamber. All operations are carried out in an inert sterile atmosphere, and all parts of the apparatus that come into contact with the milk can be sterilized.

EXPERIMENTAL

Fig. 1 is a detailed diagram of the rotary-disc apparatus. Fig. 2 shows the distillation-reconstitution apparatus and aseptic sampling chamber.

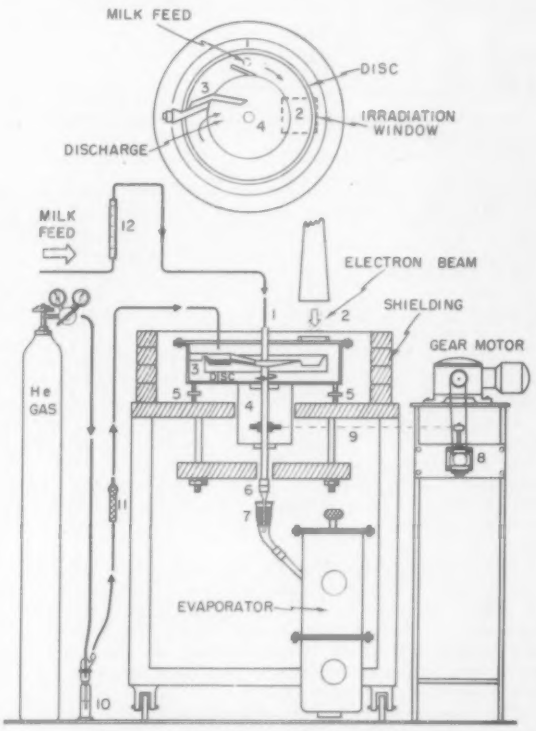

Fig. 1. Rotary-disc irradiation apparatus.

[a] Contribution Number 406 from the Department of Food Technology.

The rotating-disc apparatus (Fig. 1). The milk to be irradiated is applied by nitrogen pressure through sterile gum-rubber tubing (1), and a calibrated rotameter (12), to a rotating disc. The milk flows onto an indentation, 0.078 in. deep by 2.5 in. wide, in the outer edge of the nickel-plated aluminum disc (12-in. diameter) that rotates inside a nickel-plated brass housing. The housing is covered with a gas-tight aluminum plate having a 2 x 2-in. window (2) covered with aluminum foil (1 mil) and placed directly over the layer of milk being irradiated. During irradiation the outer surface of the window is 6 in. from the electron tube of the electron accelerator. The layer of milk is 7.5 in. from the electron tube.

The disc, rotated by a gear motor connected through a chain (9) and speed reducer (8), carries the milk through the electron beam to a flexible Tygon scraping knife (3), which causes it to flow off the disc through a nickel pipe (4), Tygon tubing (6), and sterile cotton plug (7) to the evaporator.

The irradiation chamber is maintained in a horizontal position by leveling screws (5), is well shielded with lead bricks and sheets, and is mounted on a movable cart. An inert atmosphere is maintained within the chamber by means of a constant flow of helium sterilized by passage through a wash bottle (10) containing 10% aqueous sodium hydroxide solution and a sterile cotton plug (11).

The distillation-reconstitution apparatus (Fig. 2). During irradiation the evaporator is used as a receiver for the milk. The evaporator (12 in. high and 6 in. diam), of stainless steel, is fitted with two sight glasses, a side-inlet tube and an outlet tube at the top. Its capacity is 7 L. As soon as the evaporator

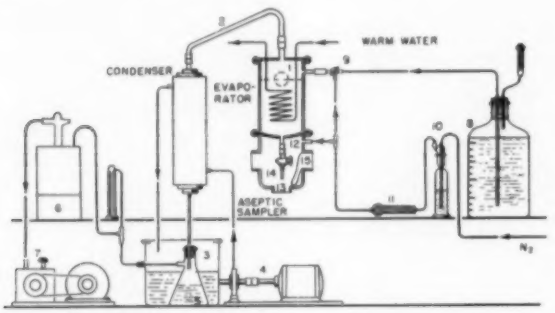

Fig. 2. Distillation-reconstitution apparatus for irradiated milk.

contains about 3 L of irradiated milk the apparatus is set up for distillation as shown in Fig. 2. Heat required for distillation is provided by a stainless-steel coil (1) through which warm water (120–130°F) is pumped. The evaporator is connected through glass tubing (2) (0.75 inch i.d.) to a shell and tube condenser fitted with a receiver (5) for the distillate immersed in an ice bath (3); to a manometer and dry-ice trap (6), and finally to a vacuum pump (7).

Evaporation is carried out at 85–90°F and 20 mm Hg pressure. The milk is maintained at constant volume during evaporation by addition of sterile water from a large reservoir (8).

Evaporation of the irradiated milk is continued until the volume of distillate collected in the receiver (5) is about 3 times the original volume of the milk being distilled. The apparatus is then brought to atmospheric pressure by admission of sterile nitrogen through a wash bottle (10) containing 10% aqueous sodium hydroxide solution and a sterile cotton plug (11).

The aseptic sampler (Fig. 2). The milk thus obtained is removed from the apparatus under sterile conditions by means of an aseptic sampling chamber. A sterile inert atmosphere is maintained in the sampling chamber by a slight positive nitrogen pressure. The milk is drained from the evaporator into sterile bottles (8 oz) through a stopcock (14), and removed with a sterile rubber glove (15).

The preparation of sterile irradiated milk. A layer (2 mm) of concentrated (4:1) milk prepared from pasteurized milk by vacuum evaporation in the Evaporator described above, was irradiated on the rotating disc in the manner outlined. The disc was rotated at 1.33 rpm (3.2 ft/min) beneath the 0.8-Mev electron accelerator, which was operated at a beam current of 0.95 ma. The practical capacity of the apparatus was 3–4 L of milk per hour.

The dose milk was exposed to had been previously determined by the methylene-blue method. Petri dishes containing 0.1% solutions (2 mm thick) of methylene blue and covered with Mylar film were irradiated on the rotating disc under the conditions given. The degree of discoloration of these solutions, as determined by comparison with a calibration curve prepared at 650 mμ with a standard Co^{60} source, indicated that the radiation dose was 1.86 megarad. This dose was shown to have a bactericidal effect in that irradiation under the above conditions of an agar growth medium inoculated with spores of a pink-pigment-producing culture of *B. subtilis*, and poured on the sterile rotating disc, resulted in no growth of the culture during storage for 3 days at 30°C.

Distillation of the irradiated milk thus collected in the evaporator was carried out at a rate of 3 L per hour. The resulting product was diluted under aseptic conditions to 3:1 concentration and stored at 30°C.

RESULTS AND DISCUSSION

Milk treated in the manner described was found to be sterile. Organoleptic evaluation in triangle tests of the freshly irradiated and distilled product indicated that it was essentially free of off-flavor.

It was well known from previous work in this laboratory that irradiated concentrated milk undergoes gelation after several days at elevated temperatures. Sterile concentrated milk (4:1) irradiated in the disc apparatus gelled in 3 days at 30°C. At low temperature (3°C), however, this milk showed no gelation after several months of storage. Gelation at 30°C could be postponed 10 days by addition of 0.2% sodium tripolyphosphate (Hoff *et al.*, 1960) to 4:1 concentrated milk before its irradiation in the disc apparatus. Gelation was postponed for at least six months by dilution of the polyphosphate-treated milk, as obtained from the apparatus, to 3:1 concentration.

The irradiated milk developed a brown color on storage at 30°C. No significant browning was observed in samples stored at 3°C. It thus appears that the irradiated milk may require refrigerated storage for higher quality.

Successful preparation of sterile irradiated milk could not be carried out unless concentrated (4:1 or 3:1) milk was used. Normal unconcentrated milk was not viscous enough to allow even distribution over the rotating disc, and thus received an uneven dose. However, use of 0.5% alginate as a thickener allowed successful preparation of sterile unconcentrated milk.

Most of the heat generated during irradiation was dissipated in the milk. Although an increase in temperature of 4°C was theoretically expected as a result of the radiation dose, the actual increase was 8–10°C. The effect, if any, of this temperature increase on the milk was not determined.

Repeated exposure of the nickel-plated aluminum disc used in the apparatus, to radiation and to milk resulted in some corrosion of the disc. It is thus clear that a stainless-steel disc is necessary if the apparatus is to be in regular use.

Acknowledgments

The authors thank the Carnation Company and the Nestlé Company, Inc., for the grants-in-aid that made this work possible. They are also grateful to the General Electric X-Ray Department for making available the 1-Mev resonant transformer used in these studies.

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Effect of Added Sugar on Consumer Acceptance of Canned Peas

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(Manuscript received December 17, 1960)

SUMMARY

Within the range 3.1–8.7% added sugar in the brine of canned peas, the highest level was preferred significantly, and the next-highest (7.35%) was preferred (both higher levels than generally used commercially). Added sugar increased total solids and alcohol-insoluble solids very slightly, gave a technically significant increase in drained weight, and appears to cause an apparent reduction in Hunter L values; there was relatively little difference between peas in the rate of change in L values of peas subjected to normal steam-table treatment.

INFORMATION is scant on the effect of added sweetness on the acceptability of canned vegetables, but recent studies indicate that consumers prefer higher levels than normally used (Buck and Weckel, 1956; Weckel *et al.*, 1960). In the continuation of studies on the effect of added sweetness on preference for canned vegetables, canned peas were studied.

METHODS

The procedure was in its entirety very similar to that used for canned whole-kernel corn (Weckel *et al.*, 1960). Peas of the variety Alsweet from one field were used to assure uniformity of quality within a pack of about 14,000 cans. The peas were packed on the regular production line of a commercial cannery in 1-lb C enamel-lined coded cans, and heat-processed 27 min at 245°F in a continuous A-B-type sterilizer. The peas were then stored 3 months at 40°F to allow equilibration. Five different concentrations of sugar in the brine ranged from 3.07% to 8.69% (Table 1). The five lots of peas were identified by code. One-half of the cans of each code were identified with a red label, and the other half with a black label.

Coded pairs of cans of peas were distributed, along with instructions for their use, and scoring forms for each member of the family 10 years of age and above to each of two groups, the Madison panel and the Kroger panel, respectively comprising 750 and 591 families. The scoring form established the following: age, sex, preference of sample for flavor and for appearance. Opportunity was provided for indicating a reason for preference of flavor (sweeter, less sweet, normal) and for appearance (brighter, darker, normal). Provision was made in the form for "no difference" observations. The principle of Latin-square arrangement was used in distribution of the peas (10 pair combinations of 5 different codes). Each family was given a red-labeled and black-labeled can in each pair, and asked to evaluate the red can first. Since each code was represented by red and black labels, half the evaluations were made in reverse order, eliminating bias from order of presentation. The Madison panel was asked not to add sugar, salt, pepper, or butter at serving the samples. The Kroger panel was allowed the option of using the condiments and butter. Both groups were asked to evaluate the samples served simultaneously with a meal of their choice. The Madison panel consisted of families whose principals were about 20% married students, 15% married office personnel, 30% university faculty, and 35% skilled and unskilled labor. Classification of the Kroger panel was not known. Responses received from the two panels totaled 10,641 score forms.

In addition, various properties of the peas were determined at intervals during storage and during steam-table treatment at 190°F for periods up to 6 hours (The Almanac of the Canning, Freezing, Preserving Industries, 1960).

Contingency tables as described by Ostle (1945) were used in analysis of the data to determine the effects of age and sex on preference. The "no difference" evaluations were excluded from the analyses.

RESULTS AND DISCUSSION

Table 1 presents the analyses of the peas at various intervals of storage and after steam-table treatments.

[a] Deceased.

Table 1. Analysis of canned peas.

	Storage period									
	72 hours					8 months				
Sugar in put-in brine (%)	3.07	4.54	5.97	7.35	8.69	3.07	4.54	5.97	7.35	8.69
Salt in put-in brine (%)	1.23	1.21	1.19	1.18	1.16	1.23	1.21	1.19	1.18	1.16
Total solids in drained peas (%)						21.6	22.7	22.9	23.6	23.8
Alcohol-insoluble solids (%)						16.3	16.3	16.7	16.9	16.8
Drained weight (g)										
8-mo. storage						309	320	329	328	331
17-mo. storage						308	314	323	322	328
pH of drained peas										
40° storage						6.33	6.30	6.32	6.23	6.25
70° storage						6.33	6.18	6.22	6.10	6.05
100° storage						5.97	5.90	5.80	5.78	5.75
Hunter "L" value										
40° storage	43.1	43.5	43.1	42.3	42.1	40.5	40.1	39.6	37.7	38.1
70° storage						38.8	38.7	39.0	37.1	37.7
100° storage	42.7	43.0	42.8	42.1	41.8	37.1	37.9	36.9	35.9	36.7
Hunter "L" value after steam-table treatment (190°F)										
0 hours						39.5	38.8	38.8	38.3	40.5
2 hours						36.3	38.1	38.1	36.5	37.0
4 hours						36.9	37.0	35.9	35.7	36.0
6 hours						36.4	36.4	35.5	35.0	35.3

Evaluation of the samples by the panels is analyzed in Table 2.

Increased levels of sugar in the put-in brine appeared to increase A.I.S. level only slightly, but increased total solids and drained weight to a greater degree. The Hunter Color Difference Meter "L" values of the peas decreased with an increase in the sugar level. The effect apparently was no greater when storage was at 100°F than at 40°F. The "L" values of the peas at each sugar level decreased during holding periods on a steam table. The rate of decrease, however, was only slightly greater for higher levels of added sugar.

Table 2. Preference evaluation of canned peas for flavor and appearance of consumer panels.

	Per cent sugar in put-in brine				
	3.07	4.54	5.97	7.35	8.69
Flavor					
Madison panel					
No. of preferences	518	907	1133	1288	1454
% of total	9.8	17.1	21.4	24.3	27.4
Kroger panel					
No. of preferences	602	890	1190	1301	1368
% of total	11.2	16.6	22.2	24.3	25.6
Appearance					
Madison panel					
No. of preferences	570	587	685	706	595
% of total	18.1	18.7	21.8	22.5	18.9
Kroger panel					
No. of preferences	559	580	746	813	668
% of total	16.6	17.2	22.1	24.1	19.8

Table 2 summarizes evaluation by the consumer panels. Higher sugar levels in the brine (7.5–8.7%) were greatly preferred. The preferred levels are at least 1.5 times those used generally by Wisconsin commercial pea canners: 1.6–5.3%, and averaging 3.25% for Alsweet (Weckel, 1960). Even higher levels of sweetness than tested might have been preferred by the panels, though there was evidence that the panels noted the adverse effect on color of the highest level of sugar. The two panels agreed remarkably in their evaluations.

A study was made of 636 interactions of sex, age, preference and reasons of preference of flavor and appearance. Only a few significant relationships were noted; these were without consistent effect. The drained weight of peas increased at least 10 g and up to 20 g per can when the sugar added per can was about 10 g. Hydration effects of the sugar were probably involved (Sterling, 1959).

Acknowledgment

Assistance of the personnel of the Oconomowoc Canning Company, Oconomowoc, Wisconsin, especially C. J. Blaska, A. J. Delwiche, and Richard Weix, is gratefully acknowledged. Funds were made available by the Sugar Research Foundation.

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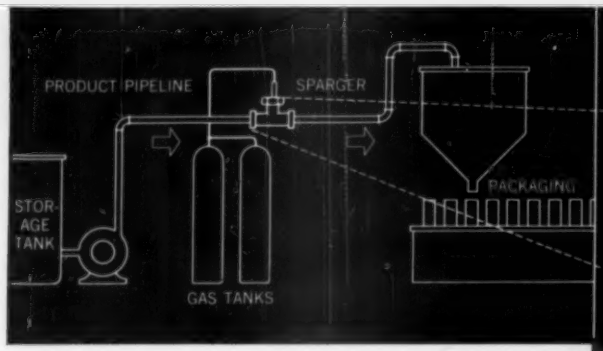

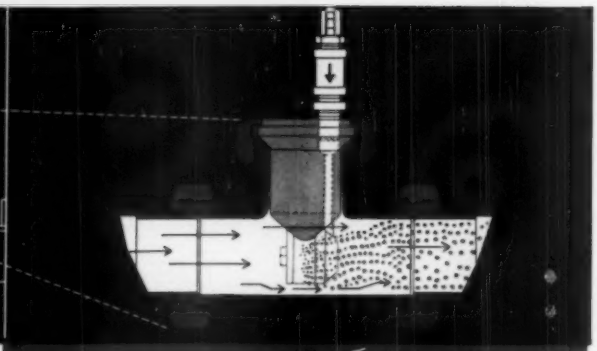

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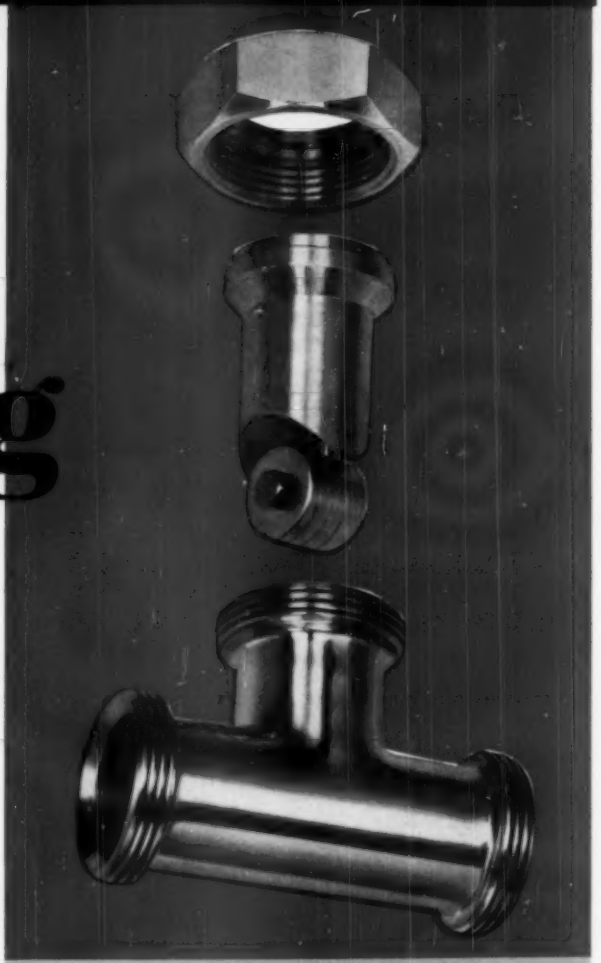

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Thermal Conductivity of Meats, Fats, Gelatin Gels, and Ice*

C. P. Lentz

(Manuscript received November 19, 1960)

SUMMARY

The thermal conductivities of ice, gelatin gels of several concentrations, different kinds of meat (including fish and poultry), and several kinds of fat, measured over a range of temperatures between 5°C and −25°C, are presented. At above-freezing temperatures the conductivities of the different meats were about equal to each other and slightly lower than the value for water. A simple linear equation expressed the relation between temperature and conductivity for the gels and meats at temperatures below −10°C. The conductivity of fats varied only slightly with temperature. Comparison of measured and calculated results indicated that calculations would be of limited usefulness in predicting the thermal conductivity of frozen or other non-homogeneous food in practice.

INFORMATION on the thermal conductivity of meats and associated animal fats is limited and frequently conflicting. Reports usually omit the composition of the sample (moisture and fat content), and frequently fail to specify test conditions such as temperature, temperature difference, and direction of grain or structure of sample with respect to direction of heat flow. In addition, few investigations have covered a range of temperatures sufficient to provide information on the variation of thermal conductivity with temperature below the freezing point. Among the exceptions are work by Cherneeva (1956) on beef, pork and associated fats, and by Jason and Long (1955) on fish. Table 1 summarizes representative published data on the thermal conductivity of meat and fat.

Knowledge of the thermal conductivity of meats and other foods is becoming more important with the present rapid increase in the amount and variety of products being frozen, particularly since this increase has been accompanied by a strong interest in faster freezing, and storage at lower temperatures. Precise and reliable data are needed on the thermal conductivity of materials to be frozen for design of equipment and predicting its performance under varying conditions, as well as for studying the effects on freezing rate of physical factors such as temperature, velocity, and properties of the heat transfer medium, and shape, size, packaging, initial temperature, and structure of the product.

The work reported here was conducted to obtain information on the thermal conductivity of commonly frozen meats and associated fats over a range of temperatures likely to be of interest in food refrigeration. Since models made of gelatin gel are frequently used in studying the effects of physical factors on cooling and freezing rates, measurements were made on gelatin gels of several concentrations. In addition, tests designed to indicate the applicability of the Maxwell-Eucken formula (Eucken, 1940; Maxwell, 1904) in predicting the thermal conductivity of non-homogeneous food materials were made with gelatin gel containing dispersed butterfat. Measurements were also made with samples of ice to establish the re-

Table 1. Values of thermal conductivity for meat and fat from published reports (values for water and ice included for comparison).

Material	Temp. (°C)	Conductivity: Cal. / (cm²)(sec)(°C/cm) × 10³
Intern. Critical Tables, 1929		
Water	20	1.40
	0	1.32
Jacob and Erk, 1929		
Ice	0	5.35
	−10	5.55
	−20	5.80
Hardy and Soderstrom, 1938		
Beef muscle	....	0.47
Beef fat	....	0.49
Awberry and Griffiths, 1933		
Beef	low	3.8
Horse meat, lean (fibers perpendicular to heat flow)	30	1.05
Whale meat (70% water)	18	0.52
Cherneeva, 1956		
Beef (fat), 74.5% water (fibers perpendicular to heat flow)	0	1.14
	−5	2.22
	−10	2.86
	−20	3.42
Beef (lean), 78.5% water (fibers perpendicular to heat flow)	0	1.14
	−5	2.53
	−10	3.22
	−20	3.75
Beef fat, 7% water	0	0.487
	−5	0.506
	−10	0.542
	−20	0.606
Pork, 76.8% water (fibers perpendicular to heat flow)	0	1.14
	−5	1.83
	−10	2.36
	−20	3.08
Pork fat, 3.1% water	0	0.444
	−5	0.542
	−10	0.606
	−20	0.695
Lapshin, 1954		
Beef fat,		
2.0% water	20	0.222
15.6% water	20	0.430
29.5% water	20	0.825
Lagoni and Merton, 1956		
Butterfat,		
0.3% water, 0.1% air	....	0.39
16.9% water, 6.5% air	....	0.47
14.5% water, 0.1% air	....	0.48
15.3% water, 10.0% air	....	0.42
Khatchaturov, 1958		
Fish (average for cod, carp, bream, pike-perch)	0	1.03
	−10	2.91
	−20	3.27
Jason and Long, 1955		
Fish (cod and haddock)	1	1.30
	−10	3.96
	−20	4.40
Smith *et al.*, 1956		
Herring (gutted, tightly packed)	−19	1.9
Cod fillets (tightly packed)	−19	2.8
Whale meat, 0.5% fat	−12	3.06

* Contribution from the Division of Applied Biology, National Research Council, Ottawa, Canada. Issued as N.R.C. No. 6281.

liability of the method of measurement and to provide a basis for comparison.

Apparatus and Procedure

The tests were made in a guarded hot-plate apparatus (Fig. 1) similar to that specified by the American Society for Testing Materials for measuring the thermal conductivity of insulating materials in slab form (ASTM Standards, 1955).[b] A number of changes were made in the standard method to adapt

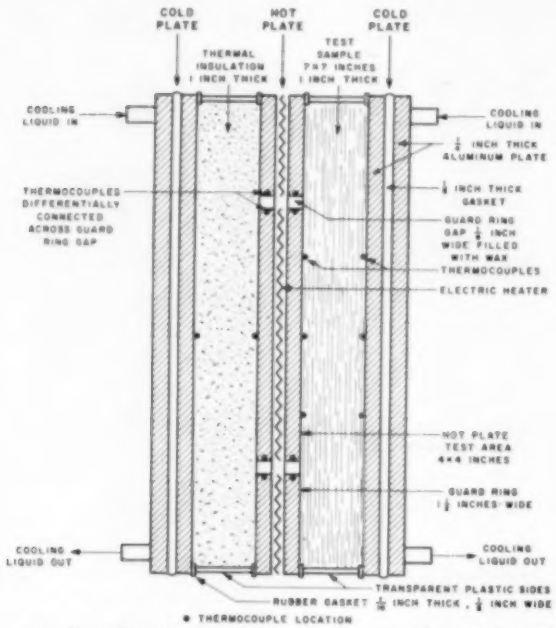
COLD PLATE
HOT PLATE
COLD PLATE
THERMAL INSULATION 1 INCH THICK
TEST SAMPLE 7×7 INCHES 1 INCH THICK
COOLING LIQUID IN
COOLING LIQUID IN
¼ INCH THICK ALUMINUM PLATE
¼ INCH THICK GASKET
THERMOCOUPLES DIFFERENTIALLY CONNECTED ACROSS GUARD RING GAP
GUARD RING GAP ¼ INCH WIDE FILLED WITH WAX
THERMOCOUPLES
ELECTRIC HEATER
HOT PLATE TEST AREA 4×4 INCHES
GUARD RING 1½ INCHES WIDE
COOLING LIQUID OUT
COOLING LIQUID OUT
TRANSPARENT PLASTIC SIDES
RUBBER GASKET ⅛ INCH THICK, ¼ INCH WIDE
● THERMOCOUPLE LOCATION

Fig. 1. Schematic sketch of hot-plate apparatus. *Notes:* 1) Dimensions in this figure are in inches; with this exception, the c.g.s. system is used throughout. 2) The hot-plate apparatus was sealed in a tank submerged in a cold bath at the sample temperature during tests.

it for use with meats, fats, and gels at low temperatures. The relatively high thermal conductivity of these materials made it possible to use insulation (of precisely known thermal conductivity) on one side of the hot plate instead of a second sample, without introducing appreciable error. This simplified the preparation of samples. Transparent plastic sides formed a leak-proof enclosure of fixed dimensions for the fluid and semi-fluid samples, and facilitated inspection of the samples. The sample-side cold plate, held in position against the edge of the plastic enclosure by clamps, could be readily removed for placing samples in the enclosure. The exterior of the hot-plate apparatus was not insulated, but errors due to fluctuations in ambient temperature were avoided by enclosing the apparatus in a container submerged in a bath controlled (±0.1°C) at about the temperature of the sample. This also reduced error in temperature measurement caused by heat conduction along thermocouple leads and prevented frost formation on the apparatus during tests at low temperatures. The cold plates were maintained uniformly at the required temperature by circulating a refrigerated alcohol solution (temperature controlled within ±0.02°C) through them at a relatively high rate. Storage batteries provided a stable source of electricity for the heaters, the power supplied to the center plate being determined with an accuracy of ±0.5%.

Temperatures were measured with butt-welded (junction only slightly larger than diameter of wire) 36-B&S-gauge copper-constantan thermocouples (locations shown in Fig. 1) used in conjunction with a potentiometer sensitive to 1 μV. Thermocouples used in balancing the guard ring heater were differentially connected across the guard ring gap, and wired directly to a sensitive galvanometer rather than to a potentiometer, to obtain a more accurate balance. All thermocouples were cemented to the plate surfaces but electrically insulated from them, and all external leads were metal-shielded to reduce stray electrical pickup.

U.S.P.-grade powdered gelatin was used in making the gelatin test samples (concentration given in per cent dry gelatin, by weight, in the gel). The gelatin gel-butterfat dispersions were mixed at 38°C rather than at room temperature so that the particles formed would be spherical rather than irregular. For the tests on meats, fish, and fats, uniform samples were selected from fresh first-grade material obtained from local retail sources.

In placing the sample material in the test apparatus, particular care was taken to prevent the formation of air spaces in the sample or adjacent to plate surfaces, since such spaces may cause serious errors. In preparing meat samples, for example, the plate surfaces to be in contact with the sample, and the pieces of meat used in making the sample, were wetted so that any spaces between pieces of sample material or between the sample and plate surfaces, would be filled with water rather than air. Meat samples prepared for testing heat flow perpendicular to the grain were made from strips of meat cut parallel to the direction of the fibers, whereas those prepared for testing heat flow parallel to the grain were made from pieces 2.54 cm (1 in.) long, cut across the fibers. In preparing samples of fat, plate and sample surfaces were "wetted" with melted fat rather than with water. The direction of the structure or grain of the fat was arranged perpendicular to the direction of heat flow. Gelatin gel samples were poured while warm through an opening in the uppermost plastic side piece.

Samples were frozen in the hot-plate apparatus itself by circulating the alcohol solution through the cold plates at −30°C. During freezing, the hot plate was heated so that a film of liquid was maintained adjacent to it until both freezing and the volume change associated with change of state and change in temperature were almost complete. The previously mentioned opening in the uppermost plastic side piece (apparatus supported on edge, as in Fig. 1) allowed sample material to be added or removed so that volume changes would not cause distortion of the frozen sample, with consequent poor contact between sample and plates. Freezing time was about the same (2–4 hours) for all samples.

In the tests made at below-freezing temperatures, a relatively small temperature difference (2–3°C) was maintained between hot and cold plates so as to minimize differences in the composition of the sample due to differences in temperature. Tests were continued until a steady-state condition was reached (1–2 days, depending on the nature of the test), and then 5–10 sets of readings were taken over 1–2 hr, depending on the steadiness of control in the particular test. These readings

[b] In the standard guarded hot-plate method, slabs of the test material of equal thickness are placed on each side of a metal plate uniformly heated by electricity (the hot plate), and a plate maintained at some temperature below the temperature of the hot plate by liquid flowing through it (cold plate) is placed in contact with the opposite side of each sample. Thermal conductivity is determined by measuring the rate of heat flow through the samples for a given difference in temperature (at least 22°C for insulating materials) between hot and cold plates. Accurate determination requires that the rate of heat flow be uniform over the test area of the sample, and in a direction normal to the face of the sample. Heat flow through the edge of the sample (edge loss) is prevented by the guard ring. The guard ring is essentially a separately heated section of the hot plate surrounding the central part of the plate, or test area, and separated from it by a gap (see Fig. 1). Heat flow to the guard ring is adjusted to where there is negligible temperature difference across the gap, and hence negligible edge loss from the test area. Insulation of the sides and edges of the apparatus reduces the effect of fluctuations in ambient temperature.

were averaged in calculating results. Preliminary tests with samples of ice showed that the apparatus would give results reproducible within ±1%.

RESULTS AND DISCUSSION

Gelatin gels and ice. Fig. 2 presents thermal conductivities over a range of temperatures for ice and for gelatin gels, with and without added sodium chloride. The curve for ice gives values slightly (about 1%) lower than those of Jacob and Erk (1929; curve shown in Fig. 2 for comparison), considered (Powell, 1958) to be the most reliable available. The curves for different concentrations of gelatin gel (without added sodium chloride) are linear and parallel to the curve for ice at temperatures below −10°C. Sodium chloride lowered the thermal conductivity of the gel markedly, presumably by reducing the amount of water frozen at temperatures above the eutectic temperature of the solution. The curves for gelatin gel with 1% and 2% added salt converge toward the eutectic point. Within the limits tested, the thermal conductivity of gelatin gels at temperatures above 0°C was independent of gelatin concentration and was only slightly higher than the values established for water.

Since the proportion of ice present in gelatin gel at a given temperature below the freezing point decreases with increasing gelatin concentration, the slope of the curves (for the gels without added salt) might be expected to decrease with increasing concentration. A progressive freezing out of ice in the sample with decreasing temperature, in amounts increasing with the initial concentration of the sample, could explain the slope of the curves, but further work would be required to confirm this. If the variation of thermal conductivity with temperature for these gels is, for the linear part of the below-freezing temperature range, expressed in the usual way:

$$K = K_o (1 + at) = K_o + K_o at$$

in which: K = thermal conductivity at temperature t
K_o = thermal conductivity at zero or reference temperature
a = coefficient of variation of thermal conductivity with temperature,

then the coefficient "a" increases with increasing gel concentration. The product $K_o a$, however, remains constant for all concentrations.

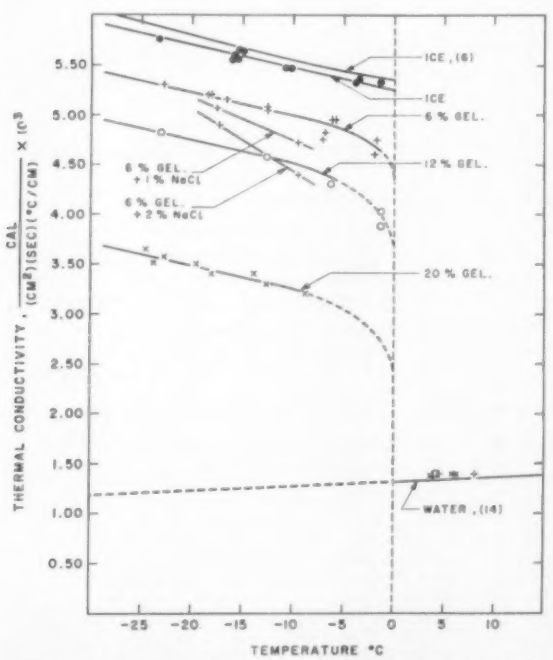

Fig. 2. Thermal conductivity of gelatin gels and ice in the temperature range 10°C to −25°C. (Concentration of gels in per cent by weight.)

Fig. 3 shows the range of thermal conductivities for mixtures of 6% gelatin gel and butterfat with the two components arranged in several different ways. Conductivity is maximum for mixtures composed of parallel layers arranged parallel to the direction of heat flow. The conductivity of this type of mixture can be calculated simply and rigorously (curve A). Conductivity is minimum, in contrast, when the heat flows perpendicular to the parallel layers (curve D). In this instance the validity of the calculation depends on the assumption of negligible contact resistance between the layers. This assumption was experimentally verified by measuring the temperatures in two adjacent layers in a sample. (There was no discontinuity at the interface.) Mixtures composed of particles of one component dispersed in the other have intermediate conductivity, as shown by curves B and C, giving calculated and measured results for small spherical particles of butterfat uniformly dispersed in the 6% gel. Comparison of the two curves shows that Eucken's adaptation (1940) of Maxwell's equation (1904) for the resistance (conductivity) of a heterogeneous substance composed of small spheres of one substance dispersed in another substance

$$K = k_c \left(\frac{1 - \left(1 - a\frac{k_d}{k_c}\right)b}{1 + (a-1)b} \right)$$

in which: K = conductivity of the mixture
k_c = conductivity of the continuous phase
k_d = conductivity of the dispersed phase

$$a = \frac{3k_c}{2k_c + k_d}$$

$$b = \frac{V_d}{V_c + V_d}$$

V_d = volume of dispersed phase
V_c = volume of continuous phase

gives results in satisfactory agreement with measured results, particularly for low proportions of the dispersed component. Since the ratio of the thermal conductivities of butterfat and 6% gelatin gel is roughly the same as for the fat and lean portions of meat, the curves of Fig. 3 give some indication of the variability that might be expected in the thermal conductivity of meat.

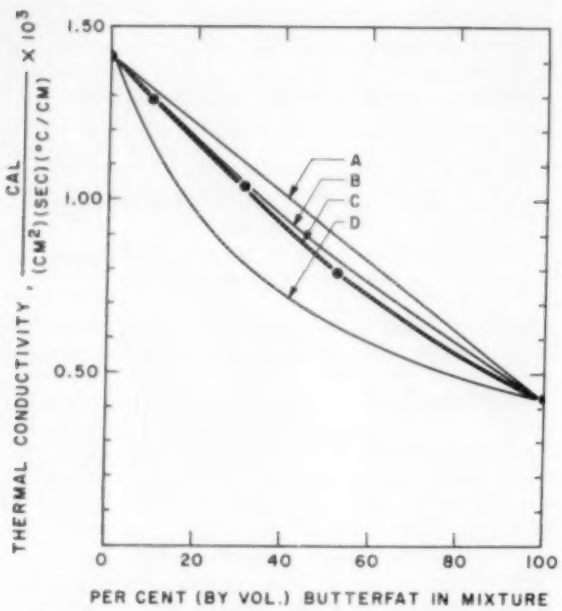

Fig. 3. Thermal conductivity of mixtures of butterfat and 6% (wt) gelatin gel at 4°C. A) Parallel alternate layers parallel to direction of heat flow (linear relation, calculated). B) Small spherical particles of butterfat dispersed in 6% (wt) gel. Values calculated from Maxwell-Eucken equation (Eucken, 1940). C) Small spherical particles of butterfat dispersed in 6% (wt) gel, experimentally determined values: particles 0.01–0.07 mm in diam for 10% (vol.) sample; particles 0.02–0.20 mm in diam for 31% (vol.) sample; particles 0.02–2.0 mm in diam for 52% (vol.) sample. D) Parallel alternate layers perpendicular to direction of heat flow (calculation based on experimentally verified assumption of negligible contact resistance between layers). *Note:* Experimentally determined values of thermal conductivity for butterfat (0.42) and 6% gel (1.41) used in calculations for curves A, B, and D.

The Maxwell-Eucken equation is sometimes used in estimating the thermal conductivity of frozen or partially frozen materials. Comparison of calculated and experimental values (Table 2) indicates good agreement for the 6% and 12% gels, and relatively poor agreement for the 20% gel. The calculations were based on the following assumptions:

1) The gels consist of ice and gel of 66% (wt) concentration at temperatures well below freezing (the linear part of the curves), as indicated by Hampton and Mennie (1934).

2) The thermal conductivity of 66% gel at below-freezing temperatures can be obtained by projecting the curve for water, as in Fig. 2.

3) Ice is the dispersed phase (spherical particles).

4) The relative volume of ice was calculated from the measured specific volume of the gel, using assumption 1, above.

The agreement for the 6% and 12% gels appears to be remarkable considering the probable error in assumption 1 and the discrepancy between assumption 3 and fact. Part of the large difference between measured and calculated values for the 20% gel may be due to the increasing error in assumption 1 with increasing concentration.

Table 2. Comparison of experimental and calculated values of thermal conductivity $\frac{\text{Cal.}}{(\text{cm}^2)(\text{sec})(°\text{C/cm})} \times 10^3$ for gelatin gels (calculated values based on the Maxwell-Eucken equation).

Gel	Temperature (°C)		
	−10	−20	−30
6% Experimental	5.00	5.22	5.45
Calculated	5.02	5.17	5.33
Difference (%)	+0.4	−1.0	−2.2
12% Experimental	4.52	4.75	4.97
Calculated	4.59	4.66	4.85
Difference (%)	+1.5	−1.9	−2.4
20% Experimental	3.75	3.48	3.72
Calculated	3.91	3.97	4.06
Difference (%)	+4.3	+14.1	+9.1

Meats and fats. Fig. 4 shows thermal conductivity

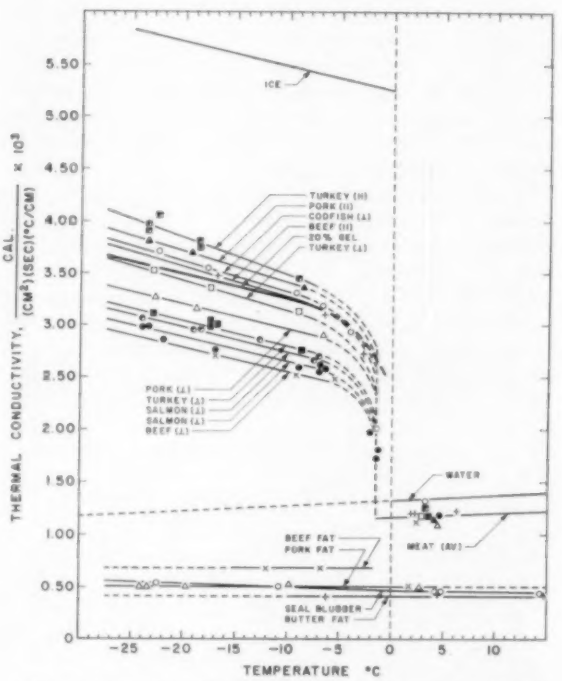

Fig. 4. Thermal conductivity of different kinds of meat, fish, and fat in the temperature range 10°C to −25°C. (Curves for ice, water, and 20% (wt) gelatin gel are shown for comparison.)

Legend (all percentages given in terms of weight):

|| Indicates test with heat flow parallel to grain of sample
⊥ Indicates test with heat flow perpendicular to grain of sample
◐ Salmon (Gaspe-Salmo salar), ⊥, 67% water, 12.0% fat
● Salmon (British Columbia—*Oncorhynchus Tshawytscha*), ⊥, 73% water, 5.4% fat
○ Codfish, ⊥, 83% water, 0.1% fat
\+ Beef (lean sirloin), ||, 75% water, 0.9% fat
× Beef (lean flank), ⊥, 74% water, 3.4% fat
▲ Pork (lean, leg), ||, 72% water, 6.1% fat
△ Pork (lean, leg), ⊥, 72% water, 6.1% fat
◩ Turkey (breast), ||, 74% water, 2.1% fat
■ Turkey (breast), ⊥, 74% water, 2.1% fat
□ Turkey (leg, ⊥, 74% water, 3.4% fat
\+ Butterfat, 0.6% water
× Beef fat (udder fat), 9% water, 89% fat
△ Pork fat (exterior), 6.0% water, 93% fat
○ Seal blubber, 4.3% water, 95% fat

measurements for different kinds of meat (including fish and poultry) and fat over a range of temperatures. Included for comparison are the curves for ice and 20% gelatin gel.

The curves for meat are all linear at temperatures below −10°C, and the slopes are equal to or slightly greater than the slope for ice and 20% gelatin gel. The equation suggested for expressing the effect of temperature on gelatin gels of various concentrations at temperatures below −10°C could apparently be used for most meats with little error. The conductivity of the sample does not appear to be directly related to moisture or fat content. It does, however, depend on the direction of the fibers or structure of the sample with respect to the direction of heat flow. Conduction was 15–30% greater along the fibers than across them. This variation, together with the relatively large differences between kinds of meat, appears to restrict the usefulness of the Maxwell-Eucken equation in predicting the thermal conductivity of meat from limited data. Eucken (1940) concluded that calculation of the thermal conductivity of mixed solids would seldom merit consideration in practice because direct measurement, although complex, is simpler than complete calculations for actual conditions. This conclusion appears justified.

At temperatures above freezing, the thermal conductivities of all the meats tested were about equal and about 10% below the established value for water.

The thermal conductivities of three of the four fats tested were about equal, and varied little with temperature. The substantially higher thermal conductivity of beef fat at below-freezing temperatures may have been due in part to its relatively high water content.

Acknowledgment

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A Method for Calculating the Temperature Distribution and Mass Average Temperature in Conduction-Heated Canned Foods During Water Cooling*

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(Manuscript received May 18, 1960)

SUMMARY

It is desirable in cooling retorted canned foods to achieve a mass average temperature between 90 and 110°F. This prevents can rusting and thermophilic spoilage. A method is described for calculating the temperature distribution curve and mass average temperature after a given cooling time has elapsed. Calculated and experimental results agreed satisfactorily. Under certain circumstances, the proposed method appears simpler to apply than existing methods.

IN THE COOLING of processed canned foods, it is usually considered desirable to cool the contents so that the mass average temperature is greater than 90°F but less than 110°F. At temperatures below 110°F thermophiles, which usually are not destroyed in processing, will not grow, and at temperatures above 90°F the surface of the can dries rapidly enough to prevent rusting.

The mass average temperature is of significance because this is approximately the temperature that the can contents assume shortly after water cooling stops. The mass average temperature then slowly decreases. The rate is slow enough to permit the possible development of thermophiles if the mass average temperature at the end of water cooling is initially too high, or to permit the rusting of cans if it is too low.

The temperature distribution during cooling may also be of interest in the evaluation of thermal processes. Herein is described a method of calculating the temperature distribution and mass average temperature in cans at the end of retort cooling.

In this study, materials that heat by conduction are considered. The problem of calculating the temperature distribution during heating has been considered by several workers, e.g., Gurney and Lurie, 1923; McAdams, 1942; Olson and Schultz, 1942; Schmidt, 1924.

In these cases the initial temperature distribution is considered uniform. In considering cooling, however, the initial temperature distribution is frequently non-uniform. If it were uniform, it would be possible to apply one of the previously cited methods for calculating the temperature distribution at the end of cooling.

Carslaw and Jaeger (1959) suggested a method that is applicable to the calculation of cooling times for an initial non-uniform temperature distribution. This solution, however, depends on the converging of a series, which is cumbersome to use under certain conditions.

Approximation methods have been suggested for calculating the temperature distribution in slabs that have, initially, a non-uniform temperature distribution (Schmidt, 1924). By applying similar techniques, it is possible to develop a method for calculating the temperature distribution and mass average temperature in a water-cooled can.

Consider a can, insulated at top and bottom by other cans, in a retort. All the heat transfer takes place through the curved surface. The mass average temperature of such a can would be the greatest of any can in the retort, and would, therefore, represent the can that would have the best temperature conditions for the development of thermophiles.

Although in normal situations no such container may exist in the retort, from the standpoint of calculation a container insulated at top and bottom by other cans suggests an extreme condition that would present favorable conditions for thermophilic spoilage, of all the cans in the retort.

Therefore, it is suggested that if the mass average temperature of this container were low enough to preclude thermophilic activity, all other cans may also be considered safe from this type of spoilage.

It is possible to calculate the temperature distribution at the end of heating by employing the dimensionless heating curves (Fig. 1) developed by Gurney and Lurie (1923). Use of these curves is illustrated in example 1:

Example 1

Determine the temperature distribution in 303 x 406 can of pea puree after heating 55 min. at 241°F in a retort. The initial temperature of the pea puree is 77°F, and the retort come-up time is negligible.

Additional Information

$$\text{Thermal diffusivity} = \frac{K}{\rho Cp} = 6.89 \times 10^{-4} \frac{(\text{sq ft})}{(\text{hr})}$$

in which K = thermal conductivity, ρ = density, Cp = specific heat.

The thermal diffusivity is essentially constant for any ma-

* Contribution Number 388 from the Department of Food Technology, Massachusetts Institute of Technology, Cambridge, Mass.

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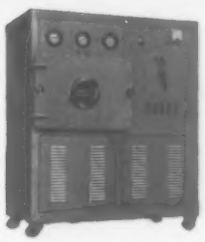

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terial. It may be determined experimentally by noting the slope of the heating curve under any conditions and applying the following equations:

if heating occurred in cylinder with ends insulated,

$$\frac{K}{\rho Cp} = \frac{r_m^2\,(.398)}{f}\left(\frac{\text{sq in.}}{\text{min.}}\right)$$

if heating occurred in cylinder with ends exposed,

$$\frac{K}{\rho Cp} = \frac{.398}{\left(\frac{1}{r_m^2}+\frac{.427}{(L/2)^2}\right)f}\left(\frac{\text{sq in.}}{\text{min.}}\right)$$

in which r_m = radius of cylinder, L = length of cylinder, f = reciprocal slope of asymptote of heating curve (Olson and Schultz, 1942). Care should be exercised to maintain a constant system of units.

Solution:

$$r_m = 1.59 \text{ in.} = 0.132 \text{ ft}$$
$$\Theta = 55 \text{ min.} = 0.916 \text{ hr}$$
$$X = \frac{K\Theta}{\rho Cp\, r_m^2} = \frac{6.98\times10^{-2}\,(.916)}{1.755\times10^{-2}} = .359 \quad \text{(See Fig. 1)}$$

Consider the radius of the cylinder divided into 6 equal parts, forming 6 annular cylinders of equal thickness (see Fig. 2).

The thickness is $\frac{r_m}{6} = \Delta R = 0.22$ ft. If distances are considered measured from the center, then the annular cylinders have the following radii:

At can surface, $r_0 = r_m = .132$ ft
$r_1 = (r_m - \Delta R) = .110$ ft
$r_2 = (r_m - 2\Delta R) = .088$ ft
$r_3 = (r_m - 3\Delta R) = .066$ ft
$r_4 = (r_m - 4\Delta R) = .044$ ft
$r_5 = (r_m - 5\Delta R) = .022$ ft
$r_6 = (r_m - 6\Delta R) = 0$

In Fig. 1, the parameter $n = \frac{r}{r_m}$ and locates the position in the container. Since the can is insulated on both ends, it behaves as an infinite cylinder, and length of the container does not influence heat transfer.

The values of "n" for the various points under consideration in the can are:

Postion	r	n
r_0	.1325 ft	1
r_1	.110 ft	.83
r_2	.088 ft	.665
r_3	.066 ft	.500
r_4	.044 ft	.334
r_5	.022 ft	.167
r_6	0	0

For these values of "n" and the value of 0.359 for "X" previously determined, it is possible to determine a value for "Y" that is related to the point temperature (see Fig. 1).

$$\text{In Fig. 1, } y = \frac{t_a - t}{t_a - t_b}$$

in which t = temperature at time Θ, t_a = ambient temperature, t_b = initial temperature.

$$\text{In Fig. 1, } m = \frac{K}{r_m h}$$

in which K = thermal conductivity and h = heat transfer coefficient at surface of container.

Since the "h" for steam will be on the order of 1000 (BTU/(hr) (sq ft) (°F) the value of "m" may be considered to be 0.

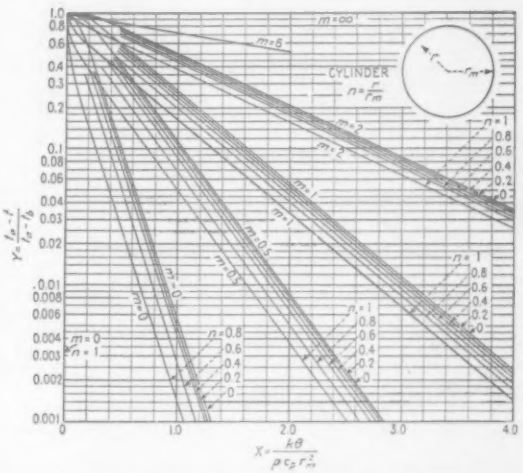

Fig. 1. Gurney-Lurie chart for infinite cylinder.

The values of "Y" and temperatures corresponding to the various values of "n" are:

Position	n	y	T_a (°F)	T_b (°F)	t (°F)
r_0	1	0	241	77	241
r_1	.83	.049	241	77	233
r_2	.665	.116	241	77	222
r_3	.500	.147	241	77	216.8
r_4	.334	.172	241	77	212.8
r_5	.167	.186	241	77	210.5
r_6	0	.189	241	77	210

A plot of the final temperature distribution after heating appears in Fig. 3.

A point .118 ft from the center was found to check experimentally with the value predicted for that position in Fig. 3.

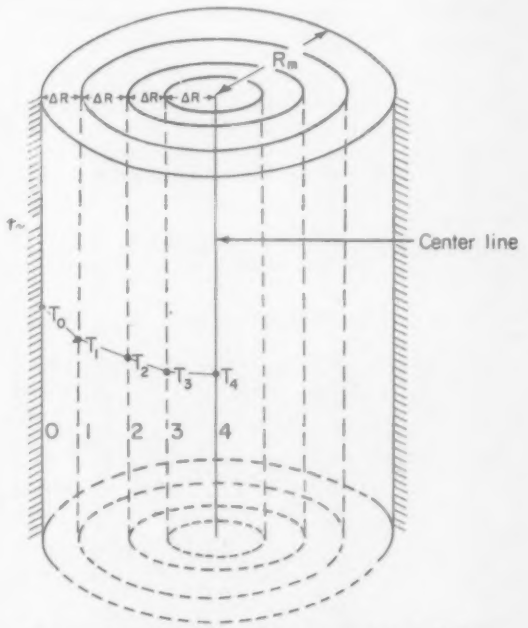

Fig. 2. Container divided into concentric cylinders.

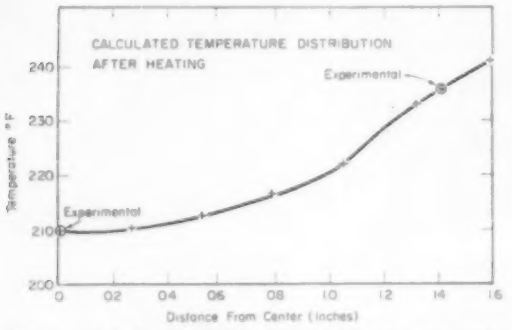

Fig. 3. Calculated temperature distribution in container after heating.

Equations for Determining Temperature Distribution in Cooling

Consider the longitudinal section of the divided cylinder in Fig. 2 (see Fig. 4). The temperature distribution through any annulus is assumed to be a straight line. It is this assumption that governs the division of the container. The slope of the line may vary with different annuli. In Fig. 4, consider a section with r_1 the center and a thickness of $\frac{1}{2}\Delta R$ on either side of r_1. A heat balance around this section, as shown in Fig 4, in an interval of time, $\Delta\Theta$, may be expressed as follows:

(Rate of heat transfer in)—(rate of heat transfer out) = (rate of heat accumulation in section) [1]

The heat accumulated in the section raises or lowers the temperature of the mass. In cooling, the heat accumulated is negative and the temperature of the mass will be decreased.

Equation (1) may be expressed mathematically as follows:

$$\frac{2\pi L\,(r_m - \tfrac{1}{2}\Delta R)\,(t_0 - t_1)K}{\Delta R} - \frac{2\pi L(r_m - \tfrac{3}{2}\Delta R)\,(t_1 - t_2)K}{\Delta R} = \frac{2\pi L\,(r_m - \Delta R)\,(\rho Cp)\,(t_1' - t_1)}{\Delta\Theta} \quad [2]$$

in which $\Delta\Theta$ = interval of time, t_0, t_1, t_2 = temperature at positions r_0, r_1, r_2, respectively, at beginning of interval of time, $\Delta\Theta$, t_1' = temperature at position r_1, at end of interval of time, $\Delta\Theta$.

Equation (2) may be simplified as follows:

$$t_1' = t_1 + \left[\frac{(r_m - \tfrac{1}{2}\Delta R)\,(t_0 - t_1) - (r_m - \tfrac{3}{2}\Delta R)\,(t_1 - t_2)}{\Delta R^2\,(r_m - \Delta R)}\right]\left[\frac{K\Delta\Theta}{\rho Cp}\right] \quad [3]$$

Considering a similar heat balance around a similar section of which r_2 is the radius, the following equation may be determined:

$$t_2' = t_2 + \left[\frac{(r_m - \tfrac{3}{2}\Delta R)\,(t_1 - t_2) - (r_m - \tfrac{5}{2}\Delta R)\,(t_2 - t_3)}{\Delta R^2\,(r_m - 2\Delta R)}\right]\left[\frac{K\Delta\Theta}{\rho Cp}\right] \quad [4]$$

Similarly, the respective equations for t_3, t_4, and t_5 are:

$$t_3' = t_3 + \left[\frac{(r_m - \tfrac{5}{2}\Delta R)\,(t_2 - t_3) - (r_m - \tfrac{7}{2}\Delta R)\,(t_3 - t_4)}{\Delta R^2\,(r_m - 3\Delta R)}\right]\left[\frac{K\Delta\Theta}{\rho Cp}\right] \quad [5]$$

$$t_4' = t_4 + \left[\frac{(r_m - \tfrac{7}{2}\Delta R)\,(t_3 - t_4) - (r_m - \tfrac{9}{2}\Delta R)\,(t_4 - t_5)}{\Delta R^2\,(r_m - 4\Delta R)}\right]\left[\frac{K\Delta\Theta}{\rho Cp}\right] \quad [6]$$

$$t_5' = t_5 + \left[\frac{(r_m - \tfrac{9}{2}\Delta R)\,(t_4 - t_5) - (r_m - 11\tfrac{7}{2}\Delta R)\,(t_5 - t_6)}{\Delta R^2\,(r_m - 5\Delta R)}\right]\left[\frac{K\Delta\Theta}{\rho Cp}\right] \quad [7]$$

The equation for the center temperature, in this case t_6', has a somewhat different form because the mass is in the form of a solid cylinder rather than an annular cylinder. Also, the heat is either being transferred in or out of the section but not both, as in the other cases.

The equation for t_6' is:

$$\frac{K2\pi\frac{\Delta R}{2}(L)\,(t_5 - t_6)}{\Delta R} = \frac{\pi\left(\frac{\Delta R}{2}\right)^2 LCp\rho\,(t_6' - t_6)}{\Delta\Theta} \quad [8]$$

Simplifying, equation (8) became:

$$t_6' = t_6 + \frac{4(t_5 - t_6)}{(\Delta R)^2}\left[\frac{K\Delta\Theta}{\rho Cp}\right] \quad [9]$$

The equation for t_0' may be determined by considering a heat balance about a section that is $\frac{1}{2}\Delta R$ thick and the outer surface of which is the surface of the container. This in effect means that the position 0 is $\frac{1}{4}\Delta R$ from the container wall. The heat balance resolves into equation (10):

$$t_0' = t_0 + \frac{h(r_m)\,(t_a - t_0) - K\,(r_m - \tfrac{1}{2}\Delta R)\,(t_0 - t_1)}{\Delta R}\left[\frac{\Delta\Theta}{\rho Cp}\right]\left[\frac{2}{r_m - \tfrac{1}{4}\Delta R}\right] \quad [10]$$

in which h = heat transfer coefficient at surface.

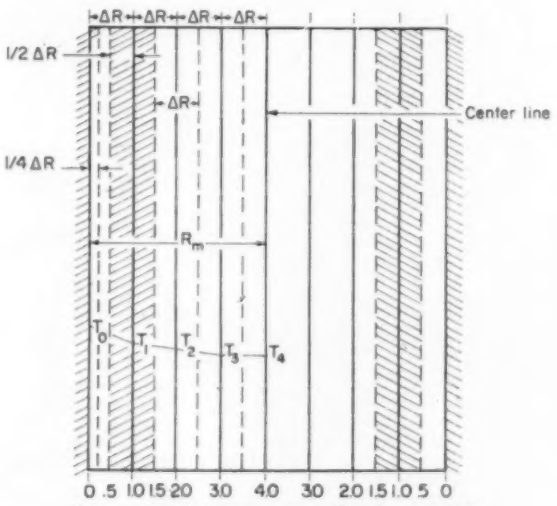

Fig. 4. Heat balance on section in container.

The heat transfer coefficient for water cooling is difficult to evaluate. Since the surface rapidly assumes the cooling water temperature, equation 10 is approximated by the following procedure:

a) At the end of the 1st interval of time, assume $t_0' = \frac{t_0 + t_a}{2}$

b) At the end of the 2nd interval of time and thereafter, assume t_0' = cooling water temperature.

This procedure was recommended by Schmidt (1924) in applying similar methods to slabs.

For cooling periods greater than 5 minutes, a time interval of one minute appears convenient. Dividing the container into 6 sections appears satisfactory. In considering very short cooling times, more divisions might be necessary for sufficient accuracy.

An illustration of how the temperature distribution curve at the end of cooling may be calculated is given in example 2.

Example 2

For the can of pea puree heated in example 1, determine the temperature distribution after cooling for 20 min. in cooling water at 77°F.

Solution:

The final temperature distribution at the end of heating becomes the initial temperature distribution of the cooling process.

In employing equations 3 to 9, many terms repeat themselves, and it is convenient to evaluate these terms separately, as follows:

$$\Delta\Theta = 1 \text{ min.} = .0167 \text{ hr}$$

$$t_a = 77°F$$

$$\Delta R = .022 \text{ ft}$$

$$\frac{K}{\rho Cp} = 6.89 \times 10^{-2} \left[\frac{\text{sq ft}}{\text{hr}}\right]$$

$$\frac{K\Delta\Theta}{\rho Cp} = 1.15 \times 10^{-4}$$

$$\Delta R^2 = 4.84 \times 10^{-4}$$

Section (1) $\Delta R^2 (r_m - \Delta R) = 5.37 \times 10^{-5}$
Section (2) $\Delta R^2 (r_m - 2\Delta R) = 4.28 \times 10^{-5}$
Section (3) $\Delta R^2 (r_m - 3\Delta R) = 3.22 \times 10^{-5}$
Section (4) $\Delta R^2 (r_m - 4\Delta R) = 2.15 \times 10^{-5}$
Section (5) $\Delta R^2 (r_m - 5\Delta R) = 1.06 \times 10^{-5}$

$(r_m - \frac{1}{2}\Delta R) = .1215$
$(r_m - \frac{3}{2}\Delta R) = .0995$
$(r_m - \frac{5}{2}\Delta R) = .0775$
$(r_m - \frac{7}{2}\Delta R) = .0555$
$(r_m - \frac{9}{2}\Delta R) = .0335$
$(r_m - \frac{11}{2}\Delta R) = .0115$

At zero time the temperature distribution is that calculated for the end of heating (see Table 1).

At the end of the first interval of time, the temperature distribution is determined with equations 3 through 9, keeping in mind the special procedure for t_o'.

For the 2nd interval of time, the temperatures at the end of the first interval serve as the initial temperatures, and equations 3 to 9 are employed. This procedure is continued until the end of the cooling period (see Table 1).

The cooling curves calculated were checked experimentally at the center and at a point 0.118 ft from the center (see Fig. 5). The experimental and calculated curves agree satisfactorily after 5 minutes of cooling. The point near the surface shows poor agreement initially because of the rapidly changing slope of the temperature distribution curve in this area. A more accurate curve could have been established by assuming smaller intervals of time and smaller cylinder divisions.

It is interesting to note that the rise in temperature experimentally observed in certain parts of the container during cooling, is predicted by this method.

Carslaw and Jaeger's Solutions for Temperature Distribution in an Infinite Cylinder

Carslaw and Jaeger (1959) described an equation for calculating the point temperature as a function of position. The method may be applied to conditions where a non-uniform temperature distribution exists. There are certain situations when this method may be more convenient to apply than the approximation method described previously, and the equation described by Carslaw and Jaeger has been discussed by Ball and Olson (1957). The point temperature may be calculated from

$$T - T_a = \frac{2}{r_m^2} \sum_{n=1}^{\infty} \frac{J_o\left[R_n \frac{r}{r_m}\right]}{J_1^2(R_n)} e^{-\left(R_n^2 \frac{K\Theta}{\rho Cp\, r_m^2}\right)} \int_0^{r_m} rf(r) \left(J_o\left(R_n \frac{r}{r_m}\right)\right) dr \qquad [19]$$

in which T_a = ambient temperature, T = temperature at r at time Θ, J_o = symbol of Bessel function of first kind of order, R_n = nth root of Bessel function of first kind of zero order, $f(r)$ = initial temperature distribution throughout container. When temperature is uniform, $f(r) = 1$.

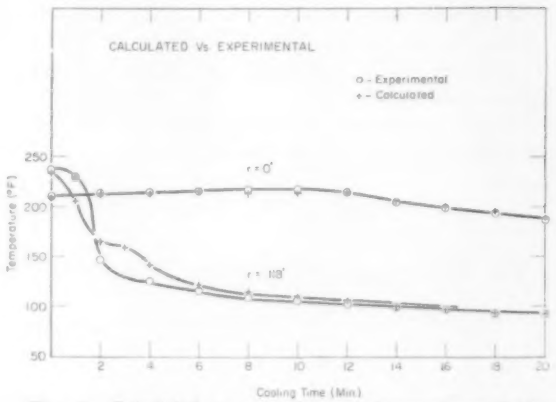

Fig. 5. Calculated vs. experimental temperatures during cooling.

Example 3

In order to illustrate the employment of equation 19, consider the problem of calculating the center-line temperature in the can described in Example 2 after 20 minutes of cooling.

r	T	T-77	rf(r)	$J_o\left[R_1 \frac{r}{rm}\right]$	$rf(r)J_o\left(R_1 \frac{r}{r_m}\right)$
0.132′	77	0	0	1	0
0.110′	233	156	17.2	0.22	3.70
0.088′	222	145	12.78	0.45	5.75
.066′	216.8	139.8	9.24	0.67	6.22
.044′	212.8	135.8	5.95	0.84	5.00
.022′	210.5	133.5	2.94	0.96	2.82
0	210.0	133.0	0	1	0

Table 1. Calculated temperature distribution (°F) during cooling at various time intervals.

$\Delta\Theta$ (hr)	t_0	t_1	t_2	t_3	t_4	t_5	t_6	t^a (r = .118′)
0	241	233	222	216.8	212.8	210.5	210.0	235.7
1/60	159	232.7	223.9	217.4	213.6	211.3	210.5	207.0
2/60	77	211.6	224.9	218.4	214.3	212.0	211.3	165.0
3/60	77	203.3	220.0	219.2	215.1	213.0	212.0	160.0
4/60	77	173.9	213.9	220.6	215.9	213.6	213.0	141
5/60	77	151.9	211.6	217.8	216.9	214.4	213.6	120.5
6/60	77	145.3	197.3	216.2	216.7	215.2	214.4	122
7/60	77	138.6	187.4	211.1	216.3	215.6	215.2	117.5
8/60	77	132.9	179.3	205.5	214.6	215.8	215.6	114
9/60	77	128.2	171.9	200.6	212.5	215.3	215.8	111
10/60	77	124.1	166.4	195.0	209.5	214.3	215.3	108
11/60	77	116.1	163.7	189.9	206.0	212.7	214.3	103
12/60	77	116.1	156.4	185.9	202.4	210.5	212.8	103
13/60	77	114.5	151.7	181.0	198.9	208.0	210.6	102
14/60	77	112.4	147.9	176.4	196.1	205.3	208.7	100
15/60	77	110.7	145.9	171.7	193.4	202.4	205.5	99
16/60	77	109.4	144.6	168.9	187.5	201.9	202.5	98
17/60	77	108.5	140.3	165.8	184.6	196.7	201.9	98
18/60	77	107.1	137.1	162.5	181.2	193.0	196.9	97
19/60	77	106.1	134.4	159.2	177.8	189.2	193.2	96
20/60	77	104.5	132.0	156.0	174.2	185 6	189.2	95

[a] $t = t_0 + (t_1 - t_0)(.66)$.

The integral is equal to 0.53.

For n = 1, the first term of the series is equal to +0.92.

For n = 2, the second term of the series, evaluated in a similar manner, is −0.0275.

For n = 3, the third term of the series is 11.9×10^{-5}.

The series converges rapidly in this case, and

$$T - 77 = \frac{2}{(.132)^2} [+.92 - 0.0275 + .00119]$$

T = 181°F.

This calculated center-line temperature compares favorably with experimental results.

In this case, the series converges rapidly enough to be practical. If this method were applied to a shorter cooling period, e.g., 5 or 10 min., the series would not converge rapidly, and many more terms in the series would have to be evaluated.

In general, the Carslaw-Jaeger solution appears to be more convenient for long cooling periods, whereas the approximation method is more convenient for shorter cooling periods.

Determination of Mass Average Temperature at the End of Heating

The mass average temperature of the container may be described mathematically as follows:

$$t_m = \frac{1}{L} \frac{[V_{(0-1)}\, t_{(0-1)} + V_{(1-2)}\, t_{(1-2)} + V_{(2-3)}\, t_{(2-3)} + V_{(3-4)}\, t_{(3-4)} + V_{(4-5)}\, t_{(4-5)} + V_{(5-6)}\, t_{(5-6)}]}{\pi r_m^2} \quad [11]$$

in which $V_{(0-1)}$ = the volume in the section between r_0 and r_1, $t_{(0-1)}$ = the mass average temperature in the section between r_0 and r_1, t_m = mass average in the container, $\pi r_m^2 L$ = the volume of container.

The temperature distribution in any section was originally assumed to be a straight line. Therefore, the temperature distribution in the section between r_0 and r_1 may be expressed by equation 12:

$$t = \frac{t_1 - t_0}{\Delta R} (r_m - r) + t_0 \quad [12]$$

The equation for determining $[t_{(0-1)}\, V_{(0-1)}] \frac{1}{L}$ may be developed as follows:

$$V_{(0-1)}\, t_{(0-1)} = \int_{\pi L(r_m - \Delta R)^2}^{\pi L r_m^2} t\,dV = L\left(\int_{r_m - \Delta R}^{r_m} \frac{t_1 - t_0}{\Delta R} (r_m - r)(2\pi r\,dr) + \int_{r_m - \Delta R}^{r_m} t_0 2\pi r\,dr\right)$$

or:

$$\frac{V_{(0-1)}\, t_{(0-1)}}{L} = \pi [r_m^2 - (r_m - \Delta R)^2]\left[\frac{t_1 - t_0}{\Delta R} r_m + t_0\right] - \left[\frac{t_1 - t_0}{\Delta R}\right]\left[\frac{2\pi}{3}\right][r_m^3 - (r_m - \Delta R)^3] \quad [13]$$

Similarly for the other sections:

$$\frac{V_{(1-2)}\, t_{(1-2)}}{L} = \pi [(r_m - \Delta R)^2 - (r_m - 2\Delta R)^2]\left[\left(\frac{t_2 - t_1}{\Delta R}\right)(r_m - \Delta R) + t_1\right] - \left[\frac{t_2 - t_1}{\Delta R}\right]\left[\frac{2\pi}{3}\right][(r_m - \Delta R)^3 - (r_m - 2\Delta R)^3] \quad [14]$$

$$\frac{V_{(2-3)}\, t_{(2-3)}}{L} = \pi [(r_m - 2\Delta R)^2 - (r_m - 3\Delta R)^2]\left[\left(\frac{t_3 - t_2}{\Delta R}\right)(r_m - 2\Delta R) + t_2\right] - \left[\frac{t_3 - t_2}{\Delta R}\right]\left[\frac{2\pi}{3}\right][(r_m - 2\Delta R)^3 - (r_m - 3\Delta R)^3] \quad [15]$$

$$\frac{V_{(3-4)}\, t_{(3-4)}}{L} = \pi [(r_m - 3\Delta R)^2 - (r_m - 4\Delta R)^2]\left[\left(\frac{t_4 - t_3}{\Delta R}\right)(r_m - 3\Delta R) + t_3\right] - \left[\frac{t_4 - t_3}{\Delta R}\right]\left[\frac{2\pi}{3}\right][(r_m - 3\Delta R)^3 - (r_m - 4\Delta R)^3] \quad [16]$$

$$\frac{V_{(4-5)}\, t_{(4-5)}}{L} = \pi [(r_m - 4\Delta R)^2 - (r_m - 5\Delta R)^2]\left[\left(\frac{t_5 - t_4}{\Delta R}\right)(r_m - 4\Delta R) + t_4\right] - \left[\frac{t_5 - t_4}{\Delta R}\right]\left[\frac{2\pi}{3}\right][(r_m - 4\Delta R)^3 - (r_m - 5\Delta R)^3] \quad [17]$$

$$\frac{V_{(5-6)}\, t_{(5-6)}}{L} = 2\pi\Delta R\left[\left(\frac{t_5 \Delta R}{2}\right) + \left(\frac{t_5 - t_6}{2}\right)(r_m) - \left(\frac{t_5 - t_6}{\Delta R}\right)(\Delta R^2)\right] \quad [18]$$

Using the values cited in Example 2, equations 13 through 18 are evaluated as follows:

$$\frac{V_{(0-1)}\, t_{(0-1)}}{L} = 1.47$$

$$\frac{V_{(1-2)}\, t_{(1-2)}}{L} = 1.62$$

$$\frac{V_{(2-3)}\, t_{(2-3)}}{L} = 1.55$$

$$\frac{V_{(3-4)}\, t_{(3-4)}}{L} = 1.29$$

$$\frac{V_{(4-5)}\, t_{(4-5)}}{L} = .817$$

$$\frac{V_{(5-6)}\, t_{(5-6)}}{L} = .317$$

Using equation 11:

$$t_m = \frac{7.07}{\pi r_m^2} = \frac{7.07}{5.53 \times 10^{-2}} = 128°F$$

Thus, the mass average temperature of the container cited in Example 2 would be 128°F after cooling for 20 minutes. Experimentally this was found to be 125°F.

Acknowledgment

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Observations on "Green" Tuna[a]

(Manuscript received November 14, 1960)

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SUMMARY

Some of the physical and chemical properties of normal and off-colored fish were compared in a shipment of tuna with a high percentage of fish that were not acceptable after being cooked. The "green" fish tended to have darker kidney suspensions, less total heme pigments, more ferri- as compared to ferroheme pigment, and higher peroxide content in the lipids. Differences in pH, total reducing substances, sulfhydryl content, total fat and TBA values were insignificant. The ranges of *all* of these measurements from fish to fish were so great as to invalidate their use as a test for greening.

In preparing tuna for canning, the eviscerated but otherwise whole fish are subjected to 3–6 hours at 215–225°F (102–107°C), allowed to cool, and trimmed to separate the loins from the heads, skin, red meat, and bones. Optimal color for the loins at this stage is a pink similar to that in canned tuna. Occasionally, however, the flesh instead is a tan to tannish-green. This phenomenon is known to the trade as "greening." The "green" fish, unsuitable for canning, are discarded. The pink has been shown to be due to a hemochrome derived from the myoglobin in the loin (Brown and Tappel, 1957), and the "green" is mostly hemichrome (Brown *et al.*, 1958; Naughton *et al.*, 1958).

If the tendency toward greening could be detected in raw fish, the waste of unnecessary handling could be avoided. Attempts to develop such a test have failed so far. Hirao *et al.* (1958a) analyzed green and normal tuna muscle and kidney tissue for a number of constituents, including B vitamins, Ca, P, and Fe, but found no usable correlation. They noted, however, that suspensions of kidney tissue from fish that later turned "green" were darker than those from fish that did not. They suggested that this difference could be used to distinguish "green" from normal tuna (Hirao *et al.*, 1958c). These differences they illustrated with a color plate (opposite page 680; Hirao *et al.*, 1958c).

During the late spring and summer of 1959, several shiploads of tuna showing an unusually high incidence of "green" fish were delivered to California canning plants. Samples were made available to this laboratory by F. E. Booth Co. The relation of kidney color to "greening" was studied, and several other analytical procedures were applied to the fish. No usable test was developed, but the observations are of interest.

EXPERIMENTAL

Samples. The fish used in this experiment were yellowfin tuna *(Neothunnus macropterus)* that the fishing company reported had been caught in the Atlantic Ocean off the coast of Africa about six weeks previously. They weighed about 80–100 lb (35–45 kg). The gills and viscera (except kidneys) had been removed, and the fish had been frozen in an air blast and held hard-frozen until delivery. In preparation for canning they had been allowed to thaw overnight in the cannery at the time our samples were taken.

Fish were selected at random and tagged. Kidney tissue (15–20 g) and tail portions (10–30 lb, 4.5–14 kg) were removed and transported to the laboratory, where the tail por-

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tions were held frozen until needed. The rest of each fish was given normal cooking in the cannery, and cooled overnight. The next morning the fish were trimmed and graded for "greenness" by qualified cannery personnel. The best fish were graded 1, and the worst, 6. Personnel from this laboratory were present at judging. Grades 5 and 6 were sufficiently off-color to be rejected for canning. Grades 1 and 2 were closest to the pink color considered normal, and grades 3 and 4 were intermediate between pink and green.

Kidney tissue. Ten g of kidney, weighed into a 100-ml beaker, with 40 ml of water, was stirred with a glass rod until the mixture was homogeneous (this pulpy tissue is readily disintegrated). The mixture was transferred to a 125-ml flask and graded for appearance. The scale was similar to that of Hirao *et al.* (1958c), but the numerical values were reversed: 1, red; 2, slightly reddish-brown; 3, reddish-brown; and 4, 5, and 6, increasingly dark to black. The graded suspensions were clarified by centrifugation for 10 minutes (4°C) at 12,000 G, and the supernatant extract was examined for visible absorption spectrum (Cary recording spectrophotometer, Model 11), pH (Leeds and Northrup, glass electrode) and apparent sulfhydryl content. The last was estimated by amperometric titration with silver nitrate according to the method of Benesch *et al.* (1955) with the modification that the titration mixture and cell were swept with oxygen-free nitrogen for 5 minutes before and during titration (Brown *et al.*, 1961).

Tuna loin extract. Thin slices of tuna meat were cut from partially thawed loins. Red meat was avoided. A 20-g sample was extracted with 20 ml of a solution of 0.001*M* EDTA. This mixture was brought to pH 7.2 by cautious addition of 2*N* sodium hydroxide, stirred with a glass rod to disperse the pieces, and allowed to stand 20–30 minutes at about 10°C. It was then poured into a gauze pad (4 layers) in a potato ricer and squeezed to remove the extract. The tissue residue was returned to the beaker and re-extracted with a second 20-ml portion of solvent. To the combined extract was added 0.4 ml of a 10% suspension of levigated alumina in distilled water. The mixture was centrifuged for 10 minutes (4°C) at 12,000 G. This clarified extract was used for determining visible absorption spectrum, sulfhydryl content (by the method described), and also the following:

Solubles. A 2-ml aliquot of the aqueous extract was dried for 18 hours at 70°C in a circulating air oven.

Volatile base. Into the outer well of a 2-in. plastic Conway diffusion cell was pipetted 1 ml of a 30% sodium hydroxide solution, and into the inner well was pipetted 4 ml of a boric acid solution containing a mixed indicator (2.5 ml of 0.1% methyl red and 8 ml of 0.1% bromocresol green per liter of 2% boric acid solution). Two ml of the aqueous extract was pipetted into the side of the outer well opposite to the alkali. The lid was placed on the rim (lightly coated with vacuum grease) and the whole cell was placed in a 50°C oven and allowed to equilibrate for about 20 minutes. The lid was resealed, and the sample was mixed with the alkali by tipping the cell in a circular pattern. After 18 hours the cell was removed from the oven and cooled, and the lid removed. The contents of the center well were titrated with 0.02*N* sulfuric acid.

Orcinol-reducing substances (pentose sugars). A 1.5-ml portion of the diluted extract (1:100) was used for crude determination of pentoses, with orcinol as the colorimetric agent (Ashwell, 1957). The standard curve was based on ribose.

Fat extraction. Fat was extracted from the tuna loin samples with chloroform-methanol according to the method of Bligh and Dyer (1959). The 50-g samples used were taken from well within the loin, i.e., at least 1 in. from the surface. Aliquots of the extract were evaporated in a stream of nitrogen for determination of yield, and other portions were taken for peroxide value (Dam and Granados, 1945).

Thiobarbituric acid assay. TBA values were determined on 5 g of minced flesh from the loin samples (Yu and Sinnhuber, 1957).

Table 1. Comparison of grade of kidney suspension to that of tuna loin.

Kidney grade[a]	No. of fish	Tuna loin grade[b]	Av.
1	9	1, 2, 2, 3, 3, 3, 4, 5, 6	3.2
2	9	1, 2, 2, 3, 3, 4, 4, 5, 6	3.3
3	5	2, 3, 3, 4, 4	3.2
4	8	1, 3, 4, 4, 4, 5, 5, 6	4.0
5	9	2, 2, 3, 3, 5, 5, 5, 6, 6	4.1
6	5	3, 3, 5, 5, 6	4.4

[a] 1, light red, to 6, black. See text.
[b] 1, pink, to 6, tannish-green. See text.

RESULTS

The grades of the cooked tuna and of the kidney suspensions (Table 1) show a trend for the "greener" fish to have darker kidney suspension, but use of this test to classify fish would lead to very large errors, at least as judged by the fish obtained in this work. On the basis of kidney grade (even ignoring the intermediate grade of 4) there would have been 15% error (4 of 23) in calling bad fish good, and a 36% error (5 of 14) in calling good fish bad.

A number of additional analytical data are presented in Tables 2, 3, and 4, as averages for each grade of cooked tuna loin. Without exception the variations from fish to fish within grade are so great as to preclude the use of any of these analytical methods as a test for "greening." Some of the trends, however, may be useful in suggesting possible causes for greening.

The pH differences are small, but appear to be slightly higher in fish that tend to green. These might be interpreted as indicating the beginning of microbiological action. The volatile bases, in contrast, have the opposite tendency, e.g., to be somewhat lower in green fish. The data therefore cannot be interpreted in terms of spoilage.

The data on orcinol-reducing substances show no trends, and the suggestion of a trend in sulfhydryl content, that is, less −SH in the greener fish, is too small to be significant in view of the wide spread of the results. The total-apparent −SH in the kidney tissue extracts (not shown) was of similar wide range (0.0–7.3m*M*/100 g) and showed a trend in the opposite direction (average 1.5m*M* in grades 1–4, average 3.1m*M* in grades 5–6).

Table 3 contains a condensation of the absorption spectra data obtained on both the loin extracts and the kidney extracts. These data suggest that total heme pigment is less in the green fish and that the pigment that is present is more likely to be in the oxidized or met-state. Again, however, the spread among the fish was too great for its use as a method of grading.

Kidney extract spectra showed a peak at 542 mμ, compared to 538 mμ for the loin. This suggests that different heme pigments are present. The pigment of tuna loin is practically all myoglobin (unpublished), but tuna kidney may contain appreciable amounts of hemoglobin.

Table 2. Tuna loin muscle analyses.[a]

Grade of cooked flesh	No. fish	pH	Volatile base (m*M*/100 g)	Reducing substances[b] (m*M*/100 g)	Water-solubles (g/100 g)	Apparent sulfhydryl (m*M*/100 g)
1–2	9	5.8(5.7–5.9)	5.6(4.5–8.1)	1.8(1.1–2.9)	10.2(5.1–14.7)	4.1(1.7–7.2)
3	11	5.8(5.7–6.1)	4.9(2.6–6.7)	1.5(0.9–1.9)	11.1(8.4–14.4)	4.1(1.3–7.8)
4	9	5.9(5.8–6.2)	4.7(2.8–6.3)	1.3(0.9–1.7)	10.8(7.5–17.4)	4.2(1.9–8.9)
5	9	6.0(5.7–6.3)	4.4(2.4–6.8)	1.4(1.1–2.3)	9.0(7.5–11.1)	3.1(1.2–5.3)
6	6	6.1(5.9–6.6)	3.9(2.2–5.7)	1.3(0.1–2.2)	8.4(5.7–10.5)	3.2(1.6–5.7)

[a] Figures in parentheses are ranges.
[b] Based on ribose.

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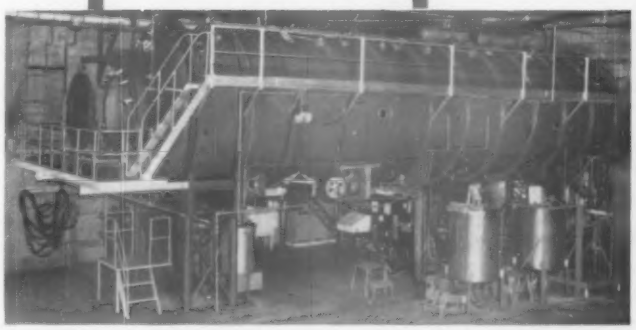

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Table 3. Absorption spectra of tuna loin and kidney.[a]

Tuna loin grades	Loins			Kidney	
	Total heme pigment[b]	538 mμ[c]	538 mμ/ 500 mμ[d]	Absorbancy 542 mμ[c]	542 mμ/ 500 mμ[d]
1–2	1.11(3)	1.78(4)	0.98(4)	1.24(8)	1.30(8)
3	1.06(6)	1.63(6)	0.94(6)	1.44(9)	1.17(9)
4	1.01(6)	1.49(5)	0.85(5)	1.32(6)	1.01(6)
5	0.77(3)	1.31(3)	0.82(3)	1.20(5)	1.20(5)
6	0.89(2)	1.31(2)	0.82(2)	1.26(3)	1.09(3)

[a] Figures in parentheses are number of fish.
[b] Absorbancy at 568 mμ of CO-treated and dithionite-reduced.
[c] A crude measure of the oxymyoglobin content (see text).
[d] A measure of the ratio of oxymyoglobin to metmyoglobin.

In the lipid analyses (Table 4), no trends were observable with total amount of lipid or TBA values but there tended to be more peroxides in the greener fish. Peroxides in the tissues could account for a subsequent oxidation of myoglobin to green decomposition products. Naughton *et al.* (1958) observed some slight correlation between peroxide content and greening.

Table 4. Lipid analyses of tuna loins.

Grade	No. of fish	Total lipid (%)	Peroxides[a]	TBA value[b]
1-2	5	0.49(0.40–0.60)	5(3–11)	57(33–89)
3	6	0.5 (0.37–0.81)	11(4–28)	59(35–104)
4	5	0.38(0.34–0.42)	15(9–23)	60(33–150)
5	9	0.43(0.30–0.58)	13(7–34)	56(27–116)
6	4	0.47(0.37–0.68)	25(8–46)	43(32–62)

[a] Millequivalents of peroxide per kg lipid.
[b] Absorbancy at 535 mμ × 100 divided by per cent lipids.

DISCUSSION

Hirao *et al.* (1958c) suggested that green fish could be easily distinguished from normal ones by the color of aqueous suspensions of kidney tissue. Our results show a general correspondence of kidney suspension color with cooked fish color, but too highly variable for use in grading.

The "greening" process appears to involve an oxidation of the normal hemochrome pigments to hemichromes (Brown *et al.*, 1958; Naughton *et al.*, 1958). It would be expected that the inherent reducing capacity of fish flesh would be one factor of importance in such cases. Some of the results with the loin extracts can be interpreted in this light. The higher percentage of oxidized (met)myoglobin in the poorer loins might be anticipated on this basis, as might the lower concentrations of sulfhydryl and reducing substances. The lower amounts of extractable pigment in the poorer fish might be due to the fact that some of the pigment proteins have been oxidized to the point of cleavage of the porphyrin ring, with consequent loss of red color. It might also be due to denaturation and resulting loss of solubility of these proteins. The latter circumstance could also explain the lower quantities of total extractable solubles from the poorer loins.

In general the peroxide value was higher for fat from the green-susceptible loins. This is in agreement with the apparent oxidative mechanism responsible for greening, since the peroxides could participate by oxidizing heme proteins. However, oxidation of the heme proteins could occur by mechanisms other than direct attack by lipid peroxides.

The results support the hypothesis that greening is an oxidative process. They afford information not previously available about raw flesh in green-susceptible loins compared to normal ones. The variability noted in every assay procedure was serious enough, however, to preclude direct development of any of the tests for prediction of tendency to greening in tuna.

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Salad Dressings Stable to Frozen Storage

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SUMMARY

Factors influencing oil and water separation from salad dressings composed of emulsions and starch paste conforming to the oil and egg-yolk specifications of the standards of identity were evaluated at temperatures ranging from +20 to −50°F. Below the freezing point (23–24°F) of the aqueous phase, 6 factors were of importance:

1) The characteristics of the oil: Oil separation is minimized when the oil used does not crystallize, crystallizes to a limited extent, or crystallizes slowly. At 20°F safflower oil is superior to other oils tested because it does not solidify at that temperature and also because good stability is achieved even though other conditions are less than optimum. When other conditions are optimum, winterized safflower oil is satisfactory at 10°F but not at 0°F or lower. With peanut oil, oil separation can be delayed for longer than 3 months at +20 to −30°F by appropriate combination of other conditions, even though this oil solidifies to some extent at +20°F. These conditions include use of the optimum thickening agent, freshly salted unfrozen egg yolk, and increase in yolk level from 5.3 to 8%, with a corresponding decrease in oil. At all storage temperatures except 0°F this combination was stable for 6 months.

2) Thickening agents: Waxy rice flour is superior to other thickening agents tested because of its slower rate of retrogradation.

3) The egg-yolk level used: Increasing it increases stability.

4) Storage condition of frozen yolk used: The emulsifying function of egg yolk decreases as the storage temperature of frozen salted yolk is lowered.

5) Salt level: Increasing it from 0.5% to 1.7% increases stability.

6) Variations in the ratio of emulsion to starch paste: Increase in the emulsion percentage increases the tendency to oil separation; increase in paste increases the tendency to water separation.

A SALAD DRESSING that can be frozen without separation on thawing has been sought by commercial producers of salads and sandwiches to make possible a greater variety of foods that can be frozen in quantity and thawed just prior to use. It has long been known that freezing usually causes the dispersed phase of emulsions to coalesce. Information on factors that contribute to the successful freezing of emulsion-containing salad dressings, obtained in the present study, should also be generally useful in developing other emulsion-containing products that would be stable if subjected to accidental or deliberate freezing.

In the standard of identity for salad dressing, the type studied is described as an "emulsified semi-solid food" prepared from edible vegetable oil, acidifying ingredients, egg-yolk-containing ingredients, a starchy paste, and optional seasoning or flavoring ingredients (U. S. Food and Drug Administration, 1950). Since emulsions and cooked starch pastes of various kinds are known to be harmed by freezing, both of these parts of the salad dressing were expected to require modification. Increased stability achieved at this laboratory by using waxy rice flour as a thickening agent for frozen sauces (Hanson *et al.*, 1951) indicated that a similar approach might increase the stability of the cooked starch paste in the salad dressing. Instability of the emulsion phase of the dressing would then be the major problem remaining.

The essential requirement of a stable emulsion is, of course, that the internal phase be prevented from coalescing. Information has accumulated in the last 40–50 years on methods of achieving this, and theories and evidence have been developed to explain the results. Egg-yolk emulsifiers function by lowering interfacial tension and rapidly forming a film around the oil globules to prevent their coalescence. The viscosity of the external phase as influenced by the emulsifying agent is considered a factor in stability; increased viscosity in the continuous phase impedes movement of the globules and thus prevents coalescence. It has been reported that if an emulsifying agent gels or produces a very viscous emulsion, the emulsion tends to be coarse but relatively stable (King and Mukherjee, 1940). Emulsions stabilized with egg yolk tend to be relatively viscous. According to Clayton (1943) the damage to emulsions that are broken by freezing is related to the influence of freezing on the emulsifying agent. He states that mayonnaise emulsions will break unless reinforced by a colloid such as pectin. Breaking of emulsions following freezing has been ascribed by Rochow and Mason (1936) to withdrawal of free and/or bound water from the films between touching globules by concentration as ice or by concentration of solutes present, followed by contact between adjacent films of emulsifiers with loss of the orienting influence of water, diffusion of emulsifier in the film away from these thick regions, and decrease in film area and coalescence of globules as soon as thawing of ice permits them to change shape.

Most studies of stability of salad dressing emulsions have primarily concerned maintenance of their stability at 35°F and above during storage, shaking, or jarring encountered in commercial marketing. Principles that apply under such conditions also apply in some respects in the +20° to −30°F range. Of primary importance is selection or treatment of the salad oil for the storage temperatures involved, and selection of an adequate emulsifying agent. Retrogradation, the tendency of starch molecules to align in a parallel manner, decreases salad-dressing stability under certain conditions. It can be prevented or

* A laboratory of the Western Utilization Research and Development Division, Agricultural Research Service, U. S. Department of Agriculture.

delayed with a suitable thickening agent. The salt level and the emulsion-paste ratio also influence stability to freezing and frozen storage.

MATERIALS AND METHODS

The formula (Table 1) was selected after preliminary testing because it conformed to the oil and egg-yolk requirements in the standard of identity of a salad dressing (U. S. Food and Drug Administration, 1950) and had a combination of stability and consistency promising enough to warrant further testing. Since the flavor and degree of stiffness would be determined by the processors, this study has been limited to the primary problem in salad dressings subjected to frozen storage, that of elimination of oil or liquid separation on thawing.

Table 1. Salad dressing formula.

Ingredients	Weight (g)	% of emulsion or paste	% of total dressing	
			Before cooking	After cooking
Starch paste				
Sugar	72	8	4.5	5.0
Waxy rice flour	99	11	6.3	6.9
5% acetic acid	225	25	14.2	44.4
Water	495.9	55.1	31.4	
Low methoxyl pectin [a]	6.75	.75	0.4	0.5
$CaCl_2$	1.35	.15	0.1	0.1
	900 g	100.0	56.9	56.9
Emulsion				
Yolk	86.8	12.4	5.3	
Salt [b]	27.3	3.9	1.7	
Sugar	34.3	4.9	2.1	
Mustard	7.0	1.0	0.4	
5% acetic acid	54.6	7.8	3.4	
Oil	490.0	70.0	30.2	
	700 g	100.0	43.1	

[a] Pectin L. M. No. 466, Exchange Lemon Products Co., Corona, California.

[b] Added in two parts, one as part of the salted yolks, remainder with dry ingredients.

Preparation of salted yolks for freezing. Unless otherwise indicated, shell eggs were stored at 35°F (2°C) until broken-out. The separated yolks were rolled on paper towels to remove extraneous white, filtered through a sieve, and blended with a wire whip in a Kitchen Aid mixer (Model G)[b] at speed No. 1; and 10% salt was added gradually during a 2-min mixing period. The bowl was scraped down, and the salted yolks were mixed for an additional 3 min. The salted yolks were poured into 8-oz glass jars (160 g/jar) and frozen.

Thawing of frozen, salted yolks. The frozen salted yolks were thawed at room temperature, 77°F (25°C), 2½ hr before using. Supplementary studies showed that results were comparable if the yolks were thawed 17–22 hours at 35°F.

Preparation of paste. Waxy rice flour was used in most cases because previous studies (Hanson *et al.*, 1951) indicated it would produce a more stable paste than ordinary cereal thickening agents. Since a paste made with this flour does not gel, a gelling agent was required to maintain the gel structure at room temperature. Low methoxyl pectin and calcium chloride were added for this purpose (Sunkist Growers Inc., 1953). In gel formation with these substances the divalent metal ions unite with carboxyls from adjacent pectinate chains, linking the chains together. When other cereal thickening agents were used, they were used at the 6.3% level in the salad-dressing formula. Water was increased from 31.4 to 31.9% to compensate for elimination of pectin and $CaCl_2$.

The dry ingredients were mixed for 10 sec with a flat beater in the Kitchen Aid mixer at speed No. 1. Water was added through the funnel attachment during the next 160 sec of mixing, and 5% acetic acid during the next 40 sec of mixing. The paste was cooked 5 min in the top of a double boiler after reaching 176°F (80°C). At the end of the cooking time the $CaCl_2$ was added as a warm solution in 20 cc of the water, and mixed for 40 sec. With thickening agents other than waxy rice flour, temperatures at the start of the 5-min cooking were 194°F (90°C) for the corn-tapioca starch mixture, and 185°F (85°C) for commercially formulated mixtures.

Preparation of emulsion. Salted yolks were blended 10 sec at speed No. 1, and ⅓ of the acetic acid was added during the next 50 sec of mixing. The sifted, dry ingredients were added gradually during the next 50 sec of mixing, and the remainder of the acetic acid during 80 sec. The oil was heated to 266°F (130°C) to destroy crystal centers, and was cooled rapidly to 86°F (30° C). Temperature of the oil pumped in the mixing bowl was 78.8°F (26°C). The oil was metered through a Zenith pump, No. 5, Graham variable-speed drive (Model 29MR5), the first ¼ being added at 28.5 ml/min, and the remainder at 57 ml/min. The emulsion was mixed for an additional 30 sec after the oil was added. At regular intervals during mixing, the bowl was scraped down with a rubber spatula.

Paste (726 g) cooled to room temperature was added to 550 g of emulsion (ratio, 1.3:1) and blended 130 sec at speed No. 1. The salad dressing was transferred to 2-oz glass jars. Within 2 hr of preparation, jars were placed in a freezing room maintained at the intended storage temperature. Time to lower the temperature from 77°F to about 2°F above the storage temperature was 3–4 hours. After storage, samples were thawed 2½ hours and examined for appearance change and separation. Separation was calculated from amount of oil separating from a 50-g sample, centrifuged 20 min in a Servall Angle Centrifuge at 1600 rpm at ambient temperature (*ca.* 77°F). Oil separation of 5–10% of the total oil present was usually accompanied by a change in appearance of the thawed dressing, characterized by a slight sponginess. Amounts of separation were greater when oil was visible in the thawed dressings before centrifuging.

RESULTS

The influence of the various ingredients on stability of salad dressings is reported in separate sections, but a general statement of the apparent over-all situation should be useful since the ingredients influence each other considerably.

In vegetable-oil salad dressings that are stable at room and refrigerator temperatures, the main cause of increasing oil separation at lower temperatures is solidification of the emulsified oil fractions in crystalline form. If an oil is used that does not solidify at the storage temperature selected (i.e., safflower oil, 20°F), no oil separation occurs. With an oil that solidifies to a limited extent at the storage temperature, oil separation can be prevented or retarded by selection of the optimum thickening agent, storage condition for egg yolk, and proportion of ingredients (i.e., egg yolk, salt) and optimum quantity of emulsion in relation to paste. Salad dressings prepared from some oils can be frozen and stored indefinitely at temperatures so low that the oils are completely solidified (i.e., −30°F) without oil separation on thawing, probably because of oil solidification in an amorphous form. Between the temperature extremes at which little or no separation occurs, there is a temperature range at which maximum oil separation occurs. In this range no combination of conditions tested delays or prevents separation adequately. Of 6 vegetable oils tested, safflower is promising because it does not solidify at 20°F (Purdy and Kneeland) and can be winterized so that it is satisfactory at 10°F in conjunction with the most stable emulsifying and thickening agents (fresh egg yolk, waxy rice flour). Peanut oil is promising because oil separation at 20 and 10° can be prevented or delayed by increasing the quantity of frozen salted yolk or using fresh salted yolks; at lower temperatures it can be delayed by increasing the quantity of fresh salted yolks.

Freezing curves for salad dressings prepared and frozen by the methods described show that the aqueous phase freezes at about 23–24°F (Fig. 1). Oil separation from the vegetable-oil salad dressings was not related to this change in the aqueous phase.

[b] Mention of commercial names of equipment and materials used does not constitute recommendation by the U. S. Department of Agriculture.

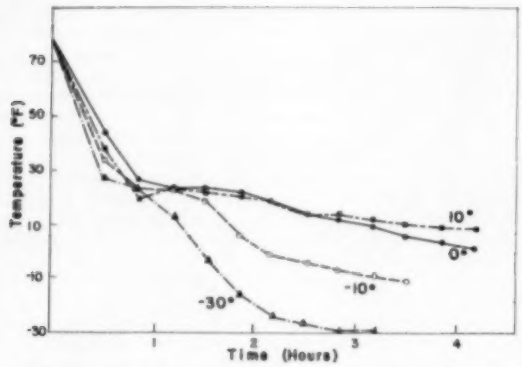


Fig. 1. Freezing curves for salad dressings held at 4 different temperatures from +10° to −30°F. The dressings were prepared from safflower oil and waxy rice flour.

Oil. The general patterns of oil separation from salad dressings stored at 77° to −50°F are similar for the vegetable oils studied, but they differ in certain respects. There was no oil separation from salad dressings held at 77 or 35°F. At successively lower temperatures, separation increased to a maximum, and then generally decreased (Fig. 2). The temperature at which oil separation is maximum varies with the oil. Two lots of cottonseed oil were used, and results from both are presented since they differ in detail though they followed the same general pattern. The first lot consistently exhibited a lesser or delayed separation at 0°F that was not typical of the second lot.

The per cent liquid glycerides in oils determined at other laboratories, and pertinent properties of the oils actually used, were tabulated (Tables 2 and 3) so that any relation between these characteristics and oil separation from salad dressings could be determined. However, according to Bailey (1950), "any method of estimating the solids and liquids in a real fat can scarcely be better than a rough approximation and can be expected to yield results more valuable for comparative than for absolute purposes." Also, the oils used in the salad dressings were used for determination of iodine value, cloud point, flow test, and titer, but not to determine per cent liquid glycerides.

There were some relationships between the oil characteristics and oil separation from the salad dressings, but they were not generally consistent enough to be useful for predicting the behavior of an oil in a salad dressing at a particular temperature. Oil separation from salad dressing generally increased as solidification of the component glycerides increased. Safflower oil, with the lowest cloud point and flow test and the highest iodine value, is the only one from which a salad dressing was made with the usual formula, that did not exhibit oil separation at 20°F. Several lots of safflower responded alike. With corn and soybean oils, which had relatively low cloud points and flow tests, and relatively high iodine values, oil separation was considerable at 20°F, although it was delayed.

Table 2. Per cent liquid glycerides in vegetable oils at temperatures from +20 to −50°F.

	Per cent liquid glycerides (approximate) at (°F):						
Oil[a]	20	10	0	−10	−20	−30	−50
Cottonseed	64	40	14	3	2	2	....
Peanut	51	18	9	5	4	2	0
Olive	....	70	40	25	18	....	....
Corn	88	78	60	49	5	0	0
Soybean	....	....	....	....	>60	52	21

[a] As reported in the references cited: cottonseed (Bailey and Oliver, 1944), peanut (Ward and Singleton, 1950), olive (Hilditch and Maddison, 1941), corn (Doerschuk and Daubert, 1948), soybean (Hilditch *et al.*, 1947).

Table 3. Properties of vegetable oils used.

Oil[a]	Iodine value[b]	Cloud point[b] (°F)	Flow test[b] (°F)	Titer[b] (°F)
Cottonseed, lot 1	114	18	16	88
Cottonseed, lot 2	114	18	18	80
Cottonseed, lot 2, winterized	114	18	16	82
Peanut	104	36	25	81
Olive	85	10	9	73
Corn	123	5	3	66
Soy bean	134	9	1	81
Safflower	144	1	0	79
Safflower, winterized	143	−17	....	82

[a] Sources of oils: cottonseed, Wesson oil, Wesson Oil and Snowdrift Sales Co., New Orleans, La.; peanut, Panther brand, Veronica Olive Oil Co., Oakland, Calif.; olive, Star brand, vergine, Olio d'Oliva product of Lucca, Italy, distr. by A. Giurlani and Bro., San Francisco; corn, Mazola, Corn Products, Argo, Ill.; soybean, Soya Oil Supreme, Durkee Famous Foods, Berkeley, Calif.; safflower, edible safflower oil, anal. 160 d 59, Pacific Vegetable Oil Corp., Richmond, Calif.

[b] Determined by A.O.C.S. method.

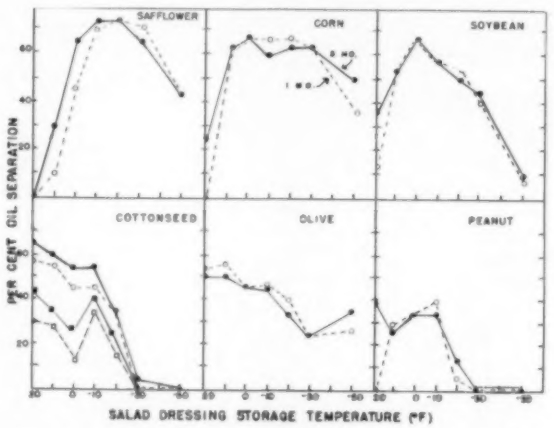


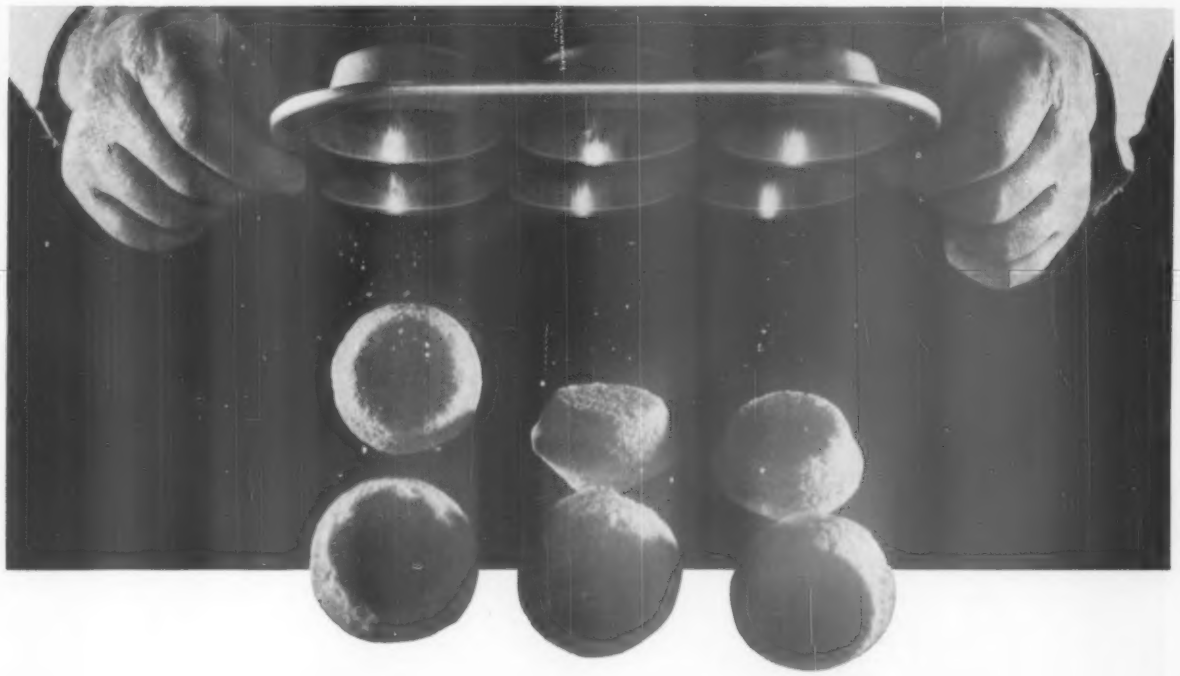
Fig. 2. Patterns of oil separation from vegetable oil salad dressings held at subfreezing temperatures for 1 and 3 months. Dressings were prepared from salted yolk held 2 months at −10°F.

On the other hand peanut oil, with the highest per cent of solid glycerides at 20°F and the highest values for cloud point and flow test, also exhibited delayed oil separation at 20°F. It is probable that the tendency of peanut oil to solidify in a "gelatinous" form (Bailey *et al.*, 1943) is largely responsible for the difference in its behavior.

At temperatures low enough to cause complete solidification of the oils, salad dressings prepared from some oils were stored indefinitely without separation on thawing, probably because solidification was in an amorphous rather than a crystalline form. Since oil solidification *per se* does not result in oil separation from vegetable oil salad dressings, the conditions that cause instability in salad dressings held at refrigerator temperature also appear to cause instability in salad dressings held at sub-freezing temperatures. Thus, extensive solidification of oil fractions in crystalline form must destroy the relation between dispersed oil droplets and the protective emulsifying film, leaving oil droplets free to coalesce on thawing. The short time (5–6 hours) required at the intermediate temperatures during freezing and thawing is apparently not sufficient for crystallization of the glycerides to occur in most cases.

Most of the oil separation occurred within the first month of storage, and little further change occurred during a year's storage. The only exceptions of importance were in the salad dressings prepared from corn, soybean, and peanut oils stored at 20°F. Although little or no separation occurred in a month's storage, significant amounts occurred after three months (Fig. 2). Other small increases on long storage occurred only under conditions that produced so much separation after one month that the changes were unimportant.

The detrimental effect on salad dressing or mayonnaise of crystallization of oil fractions at refrigerator temperature



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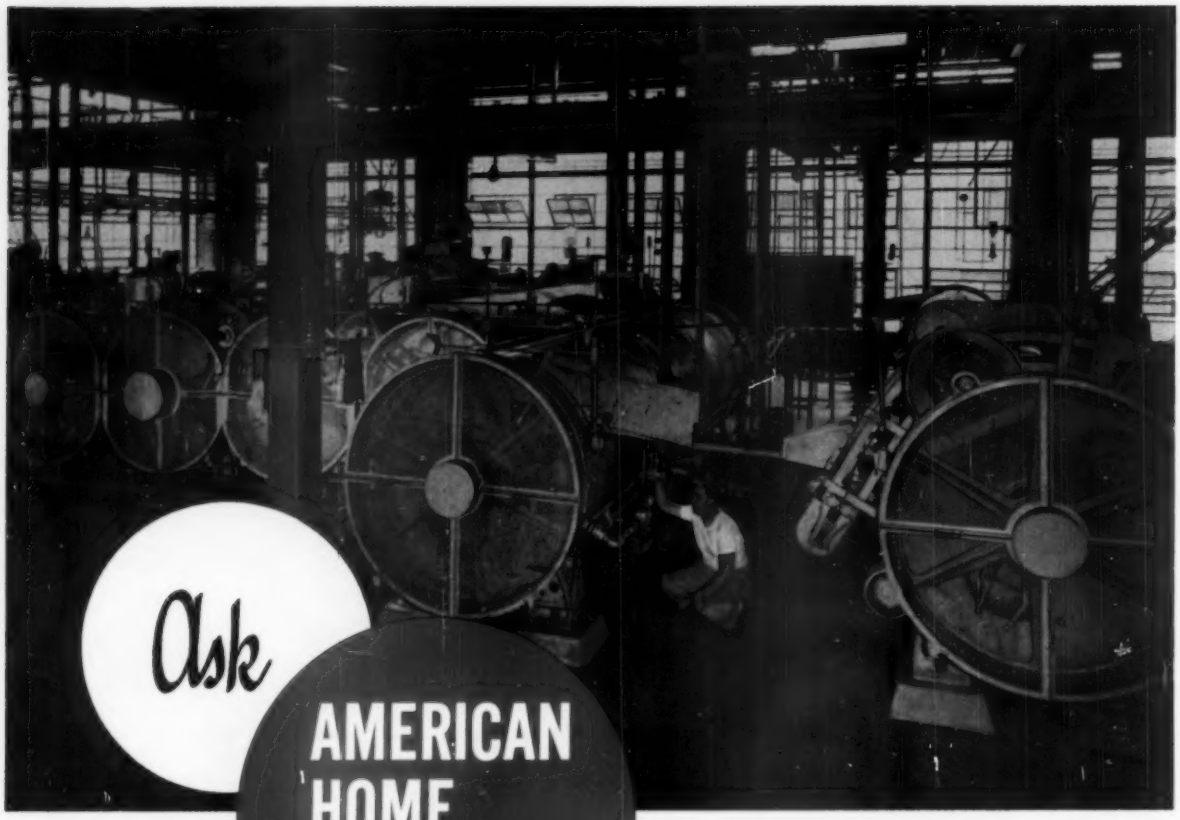

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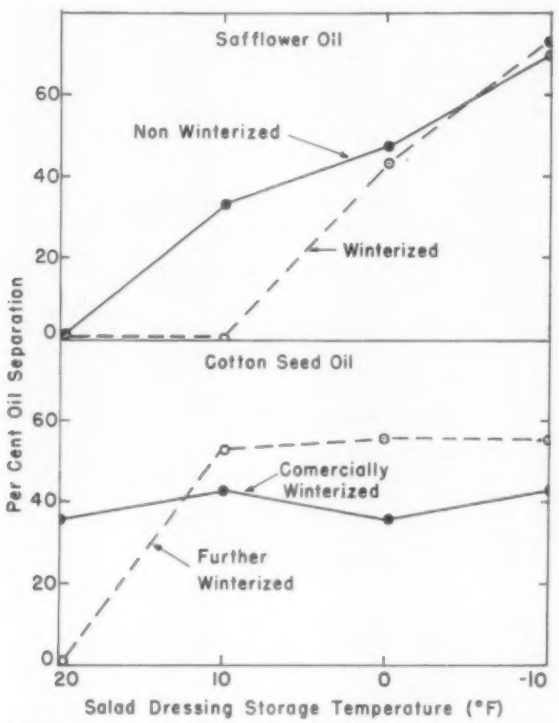

Fig. 3. Effect of "winterization" of oils on oil separation from salad dressings. The dressings were prepared from fresh salted yolk. The safflower oil dressings were stored 3 mo; cottonseed oil dressings, 1 mo.

(40–45°F) has long been recognized. Elimination or reduction of these fractions is the basis of the "winterization" of oil used in preparation of salad dressings not ordinarily subjected to freezing. Fractional crystallization of cottonseed oil at about 40°F is required. Other frequently used oils, such as corn or soybean, do not contain sufficient fractions solidifying at refrigerator temperature to require winterization. Since safflower-oil salad dressings did not separate at 20°F, this oil was selected to determine whether winterization would make the oil satisfactory for use at lower temperatures. It was centrifuged about 20 hours in a refrigerated centrifuge at 4°F. The supernatant (about 50% of the original) was slightly cloudy when decanted. However, this treatment was sufficient to prevent separation from salad dressings held 3 months at 10°F, in contrast to 33% separation from salad dressings made of untreated oil (Fig 3). Further winterization of the oil at −6°F, however, did not significantly reduce separation at 0°F.

Further winterizing of cottonseed oil that had been winterized commercially gave additional evidence that removal of fractions of the oil that solidify in a crystalline form at the storage temperature is a primary factor in stabilizing salad dressings, whether refrigerated or frozen. Removal of glycerides that crystallized at 35°F eliminated oil separation from salad dressings held 1 month at 20°F (Fig. 3).

Salad dressings made with cottonseed oil and laboratory-winterized cottonseed oil were held 1 month at low temperatures (−30 and −50°F) and then transferred to higher temperatures (+20° to −20°F) for storage to determine whether holding at the low temperature would alter or delay crystal formation and thereby reduce oil separation. Separation from the salad dressings transferred to the higher temperature equaled that of salad dressings stored for equal times at the higher temperatures without previous holding at −30 or −50°F. Similarly, when the transfer was made from the higher to the lower temperature, the separation equaled that at the higher temperature.

The maximum separation of oil from salad dressings made with the vegetable oils ranged from 40%, for peanut oil, to 73%, for safflower oil, in salad dressings containing 5.3% yolk (Fig. 2); and from 11%, for peanut oil, to 62%, for safflower oil, in dressings containing 8% yolk. These amounts were not increased by raising the temperature during centrifuging from 77 to 95°F. Peanut oil reportedly forms crystals of such "gelatinous" character that it is impossible to filter cold oil from them, and for this reason there is no commercial production of winterized peanut oil (Bailey *et al.*, 1943). It may be this characteristic of the solidified peanut oil that also prevents larger quantities of oil separation from the peanut-oil salad dressings.

Thickening agent. Oil and water separation from salad dressings prepared with four thickening agents shows that the tendency of starches to retrograde influences the stability of salad dressings subjected to frozen storage, and that the extent to which the oil solidifies or the form in which it solidifies at the storage temperature largely determines whether starch retrogradation is important. For certain conditions in which oil solidification occurs, the use of a starch that does not readily retrograde retards or prevents oil separation. Retrogradation proceeds more rapidly in unbranched than in branched starch molecules (Schoch and Elder, 1955). The common cereal starches contain 17–28% unbranched starch molecules, whereas the waxy cereal starches contain less than 1%.

The thickening agents used in the salad dressings were: 1) a 50:50 mixture of tapioca and corn starches (Purity F. L. tapioca and Kingsford corn starch), 2) waxy rice flour (plus low-methoxyl pectin and $CaCl_2$ to give necessary gel structure), 3) a commercial formulation of modified waxy and common cornstarch, and 4) a commercially formulated modified common cornstarch.

The changes in frozen salad dressing attributable to the thickening agent can be clearly shown only under conditions such that little or no oil separation occurs in salad dressings prepared with the optimum thickening agent. Under conditions resulting in large amounts of oil separation, even when the optimum thickening agent was used, no advantage was found for one thickening agent over another. The oils and the storage temperatures selected illustrate the situations existing when: 1) oil does not solidify, 2) oil solidification is slight or retarded, possibly due to crystallization in a gelatinous form, and 3) oil solidifies completely. Fresh salted egg yolks were used since studies reported later in this paper show that oil separation is less with fresh than with frozen salted yolks.

When water separation typical of starch retrogradation occurs it is visible in the centrifuged salad dressings as an intermediate layer in the aqueous phase in the centrifuge tubes. This layer was not found in salad dressings made with waxy rice flour and the standard formula, regardless of oil separation. It was found in all salad dressings made with the other thickening agents unless oil separation exceeded 40%, in which case it was probably masked by the large amounts of oil.

With any of the thickening agents tested, oil separation did not occur in salad dressings if the oils used did not contain fractions that solidify at the temperatures involved (safflower oil at 20°F) (Table 4). However, if a relatively small propor-

Table 4. Oil solidification in storage, and oil separation with various thickening agents.

	Storage temp.[a] (°F)	% oil separation[b]	
		3 cornstarch-tapioca mixtures[c]	Waxy rice flour
Not solidified:			
Safflower	20	0	0
Slight or slow solidification:			
Peanut	20	+[d]	3
Corn	20	+[d]	3
Safflower (wint.)	10	26–33	0
Complete solidification:			
Cottonseed	−50	34–46	0
Corn	−50	53–60	0
Peanut	−50	43–46	0

[a] 3 months of storage.

[b] Water separation occurred only in non-waxy rice samples at 10 and 20°F; salad dressings prepared from fresh salted egg yolks (5.3%).

[c] Cornstarch-tapioca; modified cornstarch; modified waxy and common cornstarch.

[d] Curdled, but oil not completely coalesced.

tion of the oil fractions solidify or coalescence of the oil is retarded for some other reason (corn and peanut oils at 20° and winterized safflower oil at 10°F), waxy rice flour was advantageous for a 3-month storage period. After 6 months of storage, 23% of the peanut and 12% of the corn oil separated, even with the waxy rice flour thickening agent. However, these two instances were the only ones of those reported in Table 4 in which an increase in oil separation occurred during the 3- to 6-month storage period. The gelation of the starch paste at 20°F in the three salad dressings containing cornstarch-tapioca mixtures prevented good separation of the peanut and corn oils after centrifuging, so separation is indicated as a plus rather than a number in Table 4. At a very low storage temperature (−50°F), the advantage of the waxy rice flour was again apparent.

That the greater stabilizing effect of waxy rice flour is due to the flour and not the gel-forming additives is apparent when salad dressings containing waxy rice flour, pectin, and $CaCl_2$ are compared with those containing waxy rice flour only and waxy rice flour plus pectin. In salad dressings made with cottonseed oil (Lot 1) and salted yolks (stored at −10°F), those containing waxy rice flour, pectin, and $CaCl_2$ showed oil separation of 29%, 12%, 36%, and 0% after storage for 1 month at +10, 0, −10, and −30°F, respectively. With this same combination, when waxy rice flour alone was used, there was no separation after 6 months; when waxy rice flour and pectin were used, there was no separation at +10, 0, and −30°F at 6 months, 7% after 1 month at −10°F, and 30% after 6 months at −10°F. Thus the gel-forming additives decreased the stability achieved with the flour alone. The results were similar when corn oil was used. Separation was less, though not prevented, with waxy rice flour alone than with waxy rice flour and the additives. Since waxy rice flour does not give an adequate gel structure, the additives are needed. The results suggest, however, that the starch paste gel structure is detrimental to stability, whether obtained with pectin and $CaCl_2$ or with a starch that gels.

Egg yolk. Although frozen salted egg yolks are usually used as the emulsifying agent in commercial manufacture of salad dressing, this study has shown that freshly prepared unfrozen salted yolks generally resulted in more stable frozen salad dressings. The extent of improvement depends on the storage temperature of the frozen salted yolks, and varies with oil and salad-dressing storage temperature. In general, the lower the yolk storage temperature the more the damage to its emulsifying function in salad dressing and the greater the tendency for the salad oil to coalesce on thawing. The advantage of fresh over frozen yolk is more apparent in salad dressings made with cottonseed oil (Fig. 4) than with other oils tested (Fig. 5). The increasing damage due to lower salted-yolk storage temperatures was apparent for salad dressings thickened with waxy rice flour and with the cornstarch-tapioca mixture (Fig. 6). The results in the latter tests with salted yolk held at 35°F suggest that 48 hours at that temperature has a damaging effect on the emulsifying function similar to, but less severe

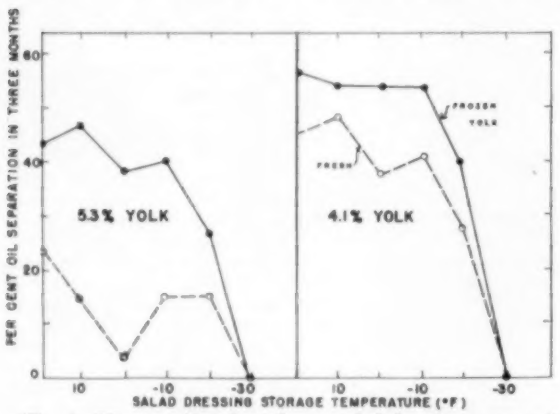

Fig. 4. Oil separation from frozen salad dressings prepared from fresh and frozen (1 month, 0°F) salted yolks at two yolk levels, and cottonseed oil.

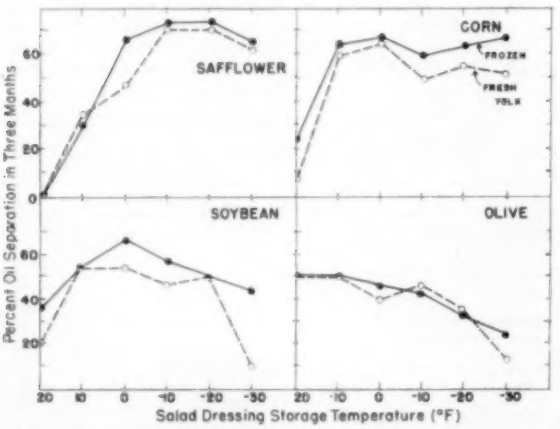

Fig. 5. Oil separation from frozen salad dressings prepared from fresh and frozen (2 months, −10°F) salted yolks and various vegetable oils.

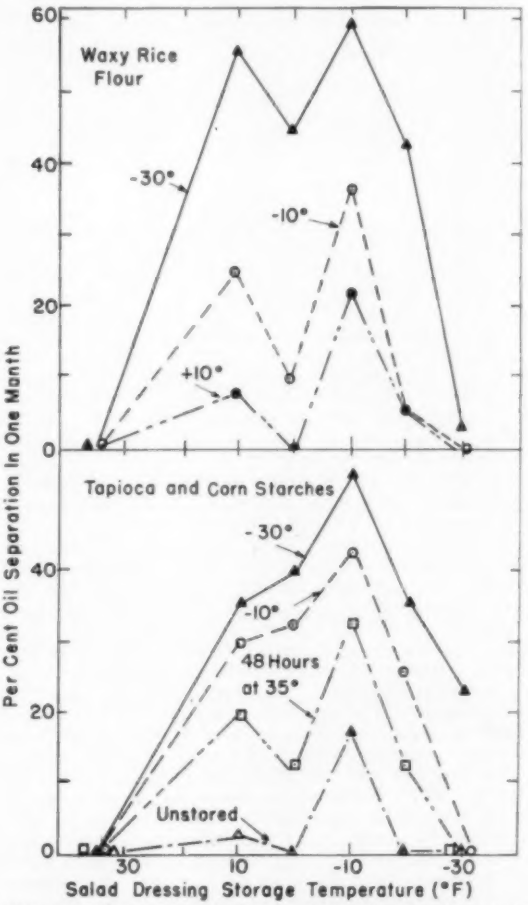

Fig. 6. Influence of salted yolk storage temperature on cottonseed oil separation from salad dressings thickened with waxy rice flour and corn-tapioca starch mixture. The salted yolks were stored 1 month.

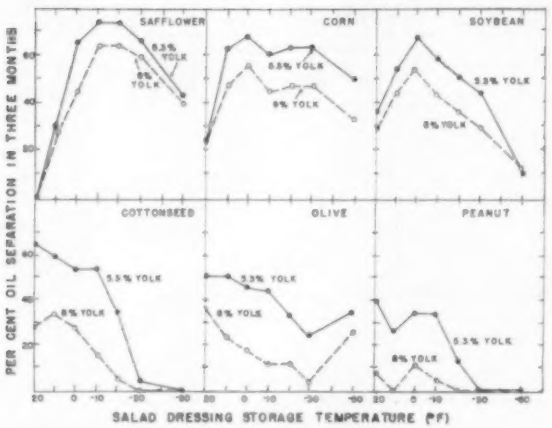

Fig. 7. Influence of yolk level on oil separation from salad dressings. The yolks were stored 2 months at −10°F.

than, that caused by frozen storage of the salted yolks. The magnitude of separation occurring with the two types of thickening agents is not directly comparable, since two egg lots were used.

Table 5. Influence of level of fresh salted yolks on oil separation from salad dressings (stored 3 months).

Oil	Yolk level (%)	20°	10°	0°	−10°
Olive	5.3	50	50	40	46
	8	29	18	15	18
Corn	5.3	7	60	63	50
	8	0	18	15	36
Soybean	5.3	20	53	53	46
	8	18	36	36	31
Safflower	5.3	0	33	46	70
	8	0	15	11	62
Cottonseed[a]	5.3	(33)	(33)	36	(26)
	8	7	(14)	4	(11)

[a] Values in parentheses indicate results after 1 mo rather than 3 mo storage.

The effect of holding temperature of shell eggs on stability of frozen salad dressings was studied at two yolk levels in salad dressings prepared from cottonseed oil. Holding shell eggs 1 week at 34°F, overnight at 55°F, or 1 week at 55°F made no statistically significant difference in oil separation from salad dressings stored 1 and 3 months at 20 to −30°F. Holding the shell eggs 1 week at 34 or 55°F before storage of the salted yolks for 1 month at 0°F likewise had no effect on the stability of salad dressings made from the same yolk levels and ingredients.

As expected, the ratio of yolk to oil in the salad dressing influences salad dressing stability. Increasing the amount of frozen salted yolk, with a corresponding decrease in oil, improves stability (Figs. 4, 7). Similar improvement is found with fresh salted yolks (Table 5). For peanut-oil salad dressings the improvement is of significant practical importance (Table 6). In these salad dressings the use of 8% fresh yolk prevents oil separation for 3 months at +20 to −30°F. Only at 0°F was there significant separation at 6 months. The increased stability is considered to be due to the resistance that the thicker film of egg surrounding each oil globule offers to the tendency of the crystallized oil to puncture the film. Joffe (1942) reported that mayonnaise containing a low per cent of winterized cottonseed oil and a high per cent (21.5%) of egg yolk could be chilled to −5°F for 24 hours without oil separation on thawing.

Salt. The salad-dressing paste, safflower-oil emulsion, and peanut-oil emulsion were stored before and after combining in the usual salad dressing formula to test the influence of salt concentration on stability. Salt increased stability of the

Table 6. Influence of level of fresh salted yolks on oil separation from peanut-oil salad dressings.

Storage temperature (°F)	Per cent oil separation					
	5.3% yolk level			8% yolk level		
	Storage time (mo.)					
	1	3	6	1	3	6
20	0	0	13	0	0	0
10	13	13	13	0	0	0
0	30	33	40	0	0	18
−10	13	13	13	0	0	2
−20	0	3	....	0	0	....
−30	0	0	....	0	0	....

frozen *pastes*. There was no water separation from the paste with a salt concentration 1.2% after 1 month at 20°F, but 18% separation from the paste with no salt. Higher salt concentrations increased stability of emulsions at 20°F (Table 7).

Table 7. Influence of salt concentration on oil separations from emulsions at 20°F (stored 3 months).

Salt concentration (%)	Solids–water ratio	Oil separation (%)[a]	
		Safflower oil	Peanut oil
1.7	1:0.88	31	70
1.7	1:1.1[b]	16	67
1.1	1:0.97	15	....
0.5	1:1.1	81	92
0.5	1:1.26[c]	81	94

[a] 70–90% separation at 10, 0, −10°.

[b] Higher solids-to-water ratio than in first sample was obtained by decreasing yolk and oil, increasing water.

[c] 1.2% sugar in this sample, 2.1% in others.

Variations in sugar concentration and in the solid-water ratio within the ranges tested had relatively little influence on emulsion stability.

In safflower or peanut oil *salad dressings* in the +20 to −10°F range, oil separation was least in those with highest total salt, whether all the salt was in the emulsion or part was in the paste (Table 8). Variations in sugar content or solids–water ratio had little if any effect on oil separation in the ranges studied. Water separation occurred only in salad dressings with the lowest (0.5%) total salt. There was no water separation from those with more salt, even though the additional salt was in the emulsion.

Emulsion—starch paste ratio. A stabilizing effect of starch pastes on the emulsions was found under some conditions. The emulsion used in this salad-dressing formula was unstable at room temperature when not combined with a starch paste. More than 50% of the oil separated from cottonseed, olive, soybean, or safflower oil emulsions after a week. Similar amounts separated when the cottonseed-oil emulsions were held for 1 month at temperatures differing by 10°F intervals from +10 to −30°F. As reported earlier in the paper, combining the emulsions with pastes made from either waxy rice flour or the cornstarch-tapioca mixture prevented oil separation from any of the salad dressings at room temperature (77°F) and decreased separation from the cottonseed oil salad dressings at −30°F.

Under some conditions a stabilizing effect of emulsions on the starch pastes was found. The conditions required to demonstrate the effect include temperatures at which water separates from starch pastes stored alone and an oil that does not solidify or solidifies to only a slight extent at those temperatures. More than 4% water separates from waxy rice flour pastes stored 1 month at 20 and 10°F, from commercially formulated modified cornstarch pastes at 20, 10, and 0°F, and from cornstarch-tapioca pastes stored from 20 to −30°F. Adding emulsions made from safflower oil or laboratory-winterized cottonseed oil to waxy rice flour pastes prevented water separation at 20°F. Adding emulsions made from winterized safflower oil prevented it at 10°F. However, the other pastes were not protected from water separation at 20°F by combining with safflower-oil emulsions or at 10°F by combining with winterized safflower-oil

Table 8. Influence of salt concentration on separation of salad dressings (stored 3 months).

Salt concentration		Sugar concentration		Solids–water ratio of emulsion	Percent oil separation							
					Storage temperature (°F)							
					Safflower oil				Peanut oil			
Emulsion (%)	Paste (%)	Emulsion (%)	Paste (%)		20	10	0	−10	20	10	0	−10
1.7	0	2.1	4.5	1:0.88	0	7	0	66	0	0	17	3
1.7	0	2.1	4.5	1:1.1	0	0	0	62	0	0	7	0
0.5	1.2	2.1	4.5	1:1.1	0	0	0	64	0	0	10	0
0.5	1.2	1.2	5.4	1:1.26	0	2	0	60	0	0	12	0
1.1	0	2.1	4.5	1:0.97	0	29	+[a]	71	....	....	....	....
0.5	0	2.1	4.5	1:1.1	0[b]	+[a, b]	6	73	0[b]	+[a, b]	13	38
0.5	0	1.2	4.5	1:1.26	0[b]	+[a, b]	16	72	0[b]	+[a, b]	12	44

[a] Oil separation occurred, but centrifuging did not separate the oil sufficiently for measurement.
[b] Water separation.

emulsions (Table 4, footnote b). These pastes were not combined with laboratory-winterized cottonseed-oil emulsions.

To determine the extent to which the ratio of emulsion to paste influence oil and water separation, salad dressings containing different ratios of emulsion to paste were prepared with safflower oil, 5.3% fresh salted yolks, and waxy rice flour. They were stored 3 months at 20°F. Water separation occurred only in the combination having a 1:4 ratio of emulsion to paste. A ratio of about 2:3 protected the paste. Oil separation occurred in the salad dressing containing 4:1 emulsion to starch paste but was prevented with 3:2 emulsion and starch paste (Table 9).

Table 9. Influence of ratio of safflower oil emulsion to waxy rice flour paste on stability of mixtures (held 3 months at 20°F).

Composition of mixture		Separation	
Emulsion	Paste	Oil	Water
%	%	%	%
0	100	0	11
20	80	0	8
42[a]	58[a]	0	0
60	40	0	0
80	20	21	0

[a] Ratio used in salad dressing.

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Stability of Chlorophyll in Vegetables as Affected by pH

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SUMMARY

Frozen green vegetables differed greatly in ability to retain chlorophyll. Destruction of chlorophyll *a* was found to be the principal factor responsible for color loss. The pH range 6–7 appeared to be critical. Experiments were carried out in which chlorophyll *a* loss in frozen green beans was retarded by cooking in water containing chemical additives. By use of citrate-phosphate buffers, pH 6.2–7.0, progressive increases in color were obtained with increase in pH. The use of buffers of pH greater than 7 resulted in marked deterioration in flavor and little further improvement in color. The buffers decreased the cooking time required for optimum texture and appeared to have no adverse effect on ascorbic acid retention.

CHLOROPHYLL retention has been used as a measure of quality in green vegetables (Dietrich *et al.*, 1957, 1959; Gilpin *et al.*, 1959; Sweeney and Martin, 1958). Color is also an important factor in consumer acceptance. Kramer estimated "eye appeal" to be about 45% of the total quality scale (Kramer and Twigg, 1956).

In a study in this laboratory of the quality of frozen vegetables purchased in the retail market it was noted that chlorophyll retention was poor in green beans and brussels sprouts. In the cooking of these vegetables a large proportion of the remaining chlorophyll was converted to pheophytin. The research reported here was undertaken to augment existing information regarding the causes of chlorophyll destruction and methods of retarding it.

Mackinney and Weast (1940) noted the effect of blanching on pheophytin formation in green beans. Dietrich *et al.* (1959) reported that blanching temperature and degree of blanch affected chlorophyll retention in frozen green beans during storage. It is also known that at an acidic pH, chlorophyll is converted to pheophytin (Willstatter and Stoll, 1928).

Previous workers attempted to prevent this conversion by addition of alkaline materials such as sodium bicarbonate, hexametaphosphate (Gilpin *et al.*, 1959), disodium glutamate, sodium hydroxide, and magnesium hydroxide (Malecki, 1958, 1959). However, because quantities required are small and waters may vary in mineral content and "hardness," the addition of these substances to the cooking water may cause a sharp increase in pH and a deterioration in texture and flavor of the vegetable. In work reported here, attempts were made to retain as much as possible of the chlorophyll in the vegetable without serious impairment of texture and flavor.

EXPERIMENTAL PLAN

Information on the stability of chlorophyll in 6 frozen vegetables, both as received and after cooking, was obtained by making determinations of total chlorophyll. The relative stabilities of chlorophyll *a* and *b* were also determined in vegetables cooked for various periods, and this stability correlated with the pH of the vegetables both before and after cooking. Attempts were then made to improve chlorophyll retention in the most acidic vegetable studied, green beans, pH about 6.2, by increasing its pH to approximately that of vegetables showing the greatest chlorophyll stability, green peas and spinach, pH 6.7–7.0. The color, flavor, and ascorbic acid retention of green beans cooked in waters buffered at pH values of pH 6.4–8.4 were also investigated.

In carrying out this work, modifications were made in the procedures for determination of the chlorophyll components, methods were improved for chromatographic separation of chlorophyll *a* and *b*, and identification was made of some of the pigments isolated by the chromatography.

MATERIALS AND METHODS

Total chlorophyll was determined in 64 samples of each of 6 frozen vegetables: broccoli spears, brussels sprouts, cut green beans, Fordhook lima beans, green peas, and spinach. Samples were collected at the retail market during 4 seasons and analyzed, both as received and after cooking.

Preparation and cooking. Seven family-size packages of each brand of the vegetable to be tested were removed from the freezer and opened. Those to be tested without cooking were allowed to thaw at room temperature for about 1 hour.

The samples to be cooked (two packages of each brand) were weighed. A predetermined amount of water (177 g for spinach, lima beans, brussels sprouts, and broccoli; 118 g for peas; 236 g for green beans) was brought to a boiling temperature as determined by thermocouple in each of four 4-quart glass-covered saucepans. The unthawed vegetable was placed in the boiling water, and timing was begun when the cooking water returned to boiling. The cooking times selected were those shown by panel evaluation to result in vegetables of near-optimum texture.

In the study of the effect of pH on color retention in frozen cut green beans, a modified cooking method was used. Samples were allowed to thaw at room temperature and 70-g portions were soaked for 10 min in the following solutions in amounts sufficient to cover the vegetable: 1) distilled water, 2) 10% monosodium glutamate (pH 6.8), 3) McIlvaine's citrate-phosphate buffer (pH 6.8). The vegetables were drained, 5 ml of the drained liquid was added to 95 ml boiling distilled water, and the sample was cooked in this liquid for 5 minutes. The color of the cooked beans was evaluated by color-difference meter, and pH measurements were taken on aqueous slurries.

A similar experiment was carried out in which samples of green beans (French style) were soaked in water, buffered with citrate-phosphate buffer at various pH's, 6.2, 6.6, and 6.9, and then cooked as described above in water to which 5 ml of the buffered solution had been added. The pH of the cooked vegetables, percentage retention of chlorophyll *a* and *b*, and color-difference meter values were determined.

In a further study of the effects of buffers ranging in pH from 6.4 to 8.4 on color, flavor, and ascorbic acid retention of green beans, both frozen and fresh samples were used. Two 10-oz packages of frozen green beans were allowed to thaw and were then immersed for 10 min in buffers, drained, and cooked until done in 225 ml boiling water to which 25 ml of the drained buffer had been added. Buffers of pH 6.4, 6.8, 7.0, 7.2, 7.6, and 8.4 were used. In addition, uncooked samples and samples cooked in water containing no added buffer were

tested. Color-difference meter and pH readings were taken. Flavor was evaluated by panel flavor scores.

One-pound samples of fresh green beans were cooked with the same procedure described for frozen vegetable. Buffers of pH 6.4, 7.0, 8,0, and 8.4 were used. Color-difference meter, pH, and ascorbic acid determinations were made on both the raw and cooked vegetables.

Flavor. Flavor of the cooked vegetable was rated by a trained panel of 5 members (Gilpin *et al.*, 1959). A five-point rating scale was used (5, denoting natural full flavor; 1, weak or lacking).

Color. Reflected color was measured with a Gardner Automatic Color Difference Meter (Gilpin *et al.*, 1959). A Gardner medium-green color standard, L 50.0, a_L–25.7, b_L + 7.3, was used. The readings were expressed as the ratio a_L/b_L.

Ascorbic acid. Reduced ascorbic acid was determined by the procedure of Bessey and King (1933). The samples were extracted according to the procedure described for broccoli (Sweeney *et al.*, 1959).

Determination of total chlorophyll. Extraction. In the extraction of chlorophyll, 100-g samples of the vegetables were blended in a Waring Blendor for 2 minutes at high speed with 100 ml of acetone. A portion of the resulting slurry (30 g for peas, green beans, and brussels sprouts, 15 g for spinach and broccoli, and 40 g for lima beans) was blended for 5 minutes with 70 ml acetone and filtered on a sintered-glass funnel, using suction. The residue was blended with 70 ml 80% acetone, filtered, and washed, and the filtrates combined and diluted to 200 ml with 80% acetone.

Because of its high chlorophyll concentration, spinach required a second dilution. A 25-ml portion of the first dilution was diluted to 100 ml with 87% acetone. For lima beans, to prevent turbidity in the extracts, a more concentrated solvent was required. Undiluted acetone (99%) was used until the final dilution, which was made with 95% acetone.

Development of absorption constants. Specific absorption constants were developed for changes in absorption that occur when chlorophyll is converted to pheophytin. This was done with freshly harvested samples of fresh vegetables assumed to contain no pheophytin. The method for development of the constants has been described (Sweeney and Martin, 1958). It should be recognized, however, that the acetone used for extraction of chlorophyll will also extract other substances, which may affect the spectral characteristics of chlorophyll. Therefore, constants developed for one vegetable might not be valid for another. In preliminary work on asparagus, determination of chlorophyll in acetone extracts was found infeasible because of interference from rutin and anthocyanin pigments. With this vegetable it was necessary, as a preliminary step, to transfer the chlorophyll to ethyl ether as described in A.O.A.C. Methods (1955).

Even in the same vegetable, constants would be influenced to some extent by differences in laboratory conditions, such as temperature, humidity, calibration of spectrophotometer, etc. Consequently it would be advisable for each analyst to develop his own constants under the conditions prevailing in his laboratory. Table 1 gives the constants developed for the 6 vegetables studied most extensively.

The method of calculating percentage retention of chlorophyll has been described (Sweeney and Martin, 1958). Briefly, the total chlorophyll plus pheophytin in mmg per ml is calculated by dividing the absorbance at 556 mμ by the specific absorption constant at that wavelength. The chlorophyll concentration is calculated from the average of the values obtained by dividing the changes in absorbance at 535, 642.5, and 660 mμ by the corresponding specific absorption constants for the changes in absorption at these respective wavelengths. The average chlorophyll value thus obtained is divided by the total chlorophyll plus pheophytin, and the quotient multiplied by 100 to give the percentage retention of chlorophyll.

Table 1. Specific absorption coefficients for conversion of chlorophyll to phetophytin in various vegetables.

Vegetable	Increase in absorbance 535 mμ	Decrease in absorbance 642.5 mμ	Decrease in absorbance 660 mμ	Absorbance 556 mμ	No. of replications
Peas	7.4	17.9	19.9	6.1	8
Broccoli	7.7	14.6	17.7	5.8	8
Brussels sprouts	8.7	13.8	16.2	7.0	6
Green beans	8.2	13.2	15.2	6.0	8
Leaf spinach	8.2	13.0	16.3	6.0	19
Fordhook lima beans	8.4	18.5	21.2	6.8	15

Determination of chlorophyll a and chlorophyll b. Determinations of chlorophyll *a* and *b* were made on uncooked frozen vegetables and on frozen vegetables cooked for 5, 10, 15, and 20 minutes.

Preparation of extracts. Acetone extracts of the vegetables were prepared as described above. Two hundred ml of extract (100 ml for broccoli, 50 ml for spinach) was then transferred to petroleum ether and washed by repeated bubbling through water (Assoc. of Offic. Agr. Chemists, 1955). Six washings were sufficient for broccoli and spinach; 10 were required for peas, green beans, lima beans, and brussels sprouts. The acetone-free extract was transferred to a 50-ml volumetric and dried over anhydrous sodium sulfate. A 10-ml portion was then chromatographed.

Chromatographic procedure. Chromatography was carried out on powdered sugar. Some modifications were made in the previously described procedure (Sweeney and Martin, 1958). The column was washed with petroleum ether, and after addition of the sample, elution was begun with petroleum ether containing 5% acetone.

After the chlorophyll bands had separated on the column, an eluting solution containing 10% acetone in petroleum ether was used until the carotene, pheophytin *a*, and xanthophylls had been eluted. Wider separation of bands was achieved by reducing the rate of flow to no more than 50 drops per minute.

After chlorophyll *a* had been eluted, chlorophyll *b* could be removed from the column by using a stronger eluting solution, 15–25% acetone in petroleum ether.

An ultraviolet lamp can be used to follow the progress of the bands. The chlorophylls and pheophytins have a red fluorescence in u.v. light, whereas the carotenes and xanthophylls show no fluorescence.

The first pigment to be eluted from the column was in the form of a diffuse yellow band that was identified as a carotene. In vegetables that had been cooked for 20 minutes, the following order of elution was obtained: diffuse yellow (carotene), gray band (pheophytin *a*), 2 yellow bands (xanthophyll), blue green (chlorophyll *a*), yellow green (chlorophyll *b*), yellow (xanthophyll, obtained only in lima beans, spinach, and green beans). In two vegetables, brussels sprouts and green peas, a second gray band (pheophytin *b*) was observed following chlorophyll *b*.

The various bands eluted from the column were identified by their spectral (Assoc. of Vitamin Chemists, 1951) and solubility characteristics (Karrer, 1950).

The fractions containing the chlorophylls were each diluted to 50 ml with petroleum ether. To one 25-ml aliquot was added 1 ml of saturated oxalic acid in acetone; to the other, 1 ml of acetone. After 30 minutes, absorbance readings were taken at 535, 556, 600, 642.5, and 660 mμ.

Calculation of results. It was determined that:

$$\frac{\text{absorbance in ethyl ether}}{\text{absorbance in petroleum ether + 10\% acetone}} = 1.29.$$

Using this information and the specific absorption constants listed in A.O.A.C. (1951) for chlorophylls *a* and *b*, the following constants were calculated:

Wavelength (mμ)	Specific absorption constants in petroleum ether containing 10% acetone: chlorophyll *a*	chlorophyll *b*
600	7.7	.....
642.5	12.6	44.6
660	79.1	7.7

These constants were used to calculate the amounts of chlorophyll *a* and *b* in chromatographed extracts of the uncooked vegetables. Constants for changes in absorbance upon conver-

sion of the chlorophylls were then calculated, using the same method as for total chlorophyll (Sweeney and Martin, 1958). These constants were used for calculation of the amounts of the chlorophylls and pheophytins in chromatographed extracts of the cooked vegetable.

RESULTS

Total chlorophyll. Table 2 shows data on mean values for percentage retention of chlorophyll in 6 vegetables. Also shown are pH values obtained on the cooked and uncooked vegetables. In general, vegetables showing the highest percentage retention as received, also had the best chlorophyll retention when cooked. As would be expected, there was a relationship between pH and chlorophyll stability since spinach and green peas, which were best in chlorophyll retention, also had the highest pH. Conversely, brussels sprouts and green beans, which were lowest in pH when cooked, were also lowest in percentage retention of chlorophyll.

Table 2. Percentage retention of chlorophyll in frozen vegetables as affected by pH.

Vegetables	Uncooked		Cooked [a]	
	Chlorophyll retention [b] %	pH [c]	Chlorophyll retention [b] %	pH [c]
Spinach	90.0	6.8	77.2	6.7
Peas	89.8	7.0	67.6	6.7
Broccoli	81.4	6.5	44.9	6.4
Fordhook lima beans	72.5	6.5	40.1	6.5
Green beans	76.6	6.2	26.7	6.0
Brussels sprouts	69.0	6.3	20.7	6.2

[a] The cooking time in each case was that required for optimum texture.
[b] Mean of 64 samples.
[c] Mean of 4 samples for all vegetables except green beans; values for green beans, mean of 10 samples.

Chlorophyll a and b. In Fig. 1, the percentages of chlorophyll retained as chlorophyll *a* are plotted against cooking time. In general, results are similar to those obtained with total chlorophyll. Spinach and green peas, high in retention of total chlorophyll, were also highest in the percentage retained as chlorophyll *a*. Green beans and brussels sprouts, again, were lowest.

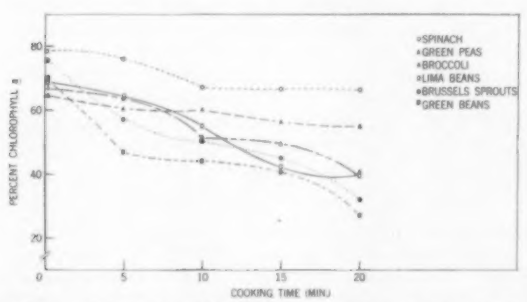

Fig. 1. Effect of cooking time on retention of chlorophyll a.

The results indicate that destruction of chlorophyll *a* was the principal factor responsible for loss of color in cooked green vegetables. Table 3 shows values for the percentage retention of chlorophyll *a* and chlorophyll *b* in broccoli cooked for various periods. The greater stability of chlorophyll *b* than of chlorophyll *a* is evident.

Table 3. Retention of chlorophyll *a* and chlorophyll *b* in cooked broccoli.

Cooking time (min)	% Chlorophyll retention		
	Chlorophyll	Chlorophyll *a*	Chlorophyll *b*
5	82.5	78.7	90.3
10	58.9	45.2	86.7
15	47.0	35.1	71.2
20	31.3	18.4	57.6

Since there were striking differences in chlorophyll *a* stability in vegetables such as spinach or green peas, pH 6.7–7.0, as compared to brussels sprouts and green beans, pH 6.0–6.3, it appeared that the pH range between about 6 and 7 might be critical in maintaining the stability of chlorophyll *a*.

This relationship was therefore investigated further. Samples of frozen green beans were cooked for various periods in water containing no additive, and in water to which either monosodium glutamate or citrate-phosphate buffer had been added for control of pH. The cooking methods used are described above.

Effect of pH adjustment on color retention. Fig. 2 shows color-difference meter readings on cut green beans cooked in water containing additives for control of pH. Comparisons are made with the color of uncooked green beans and green beans cooked in water containing no additive.

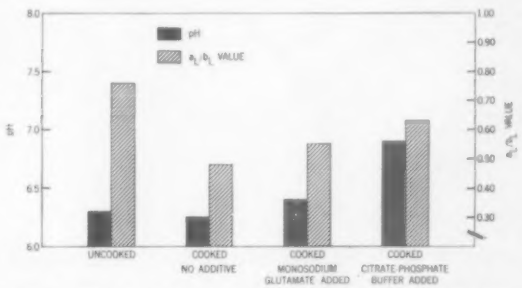

Fig. 2. Effect of chemical additives on color retention in cut green beans when cooked.

The use of monosodium glutamate caused some increase in pH and color-difference meter a_L/b_L values. The citrate-phosphate buffer caused a greater increase in both pH and a_L/b_L values.

Fig. 3 shows the effect of pH on chlorophyll *a* retention and a_L/b_L values of French-style green beans. The pH values shown, 6.4, 6.9, and 7.0, are values obtained on vegetables cooked in liquids buffered respectively at 6.2, 6.6, and 6.9. Retention of color in cooked green beans improved with each increase in pH of the buffer. This is indicated by higher retention of chlorophyll *a* (50% in green beans cooked in water buffered at pH 6.9, compared to 35.5% in water containing no additive) and by a corresponding increase in a_L/b_L values, from 0.49 to 0.71.

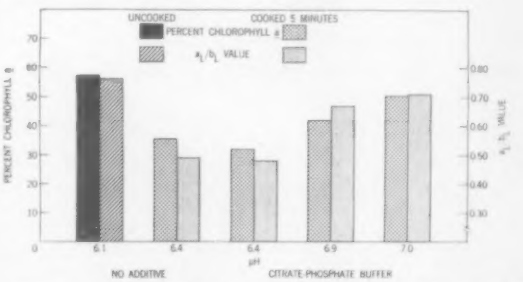

Fig. 3. Effect of pH adjustment on retention of chlorophyll a in frozen green beans when cooked.

Fig. 4 shows color-difference meter a_L/b_L values and panel flavor scores obtained on frozen green beans cooked in water buffered over the range pH 6.4 to 8.4. Buffers of pH greater than 7 resulted in little further color improvement and produced a sharp decrease in panel flavor scores.

Fig. 5 shows color and ascorbic acid determinations made on fresh green beans cooked in liquids buffered over the range

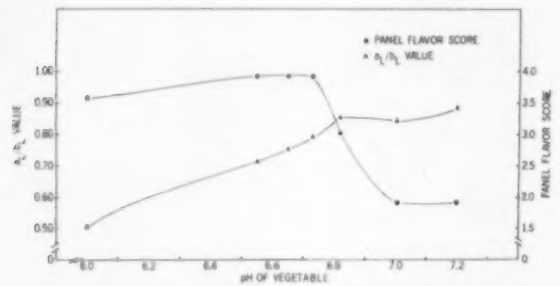

Fig. 4. Effect of pH adjustment on color and flavor of frozen green beans when cooked.

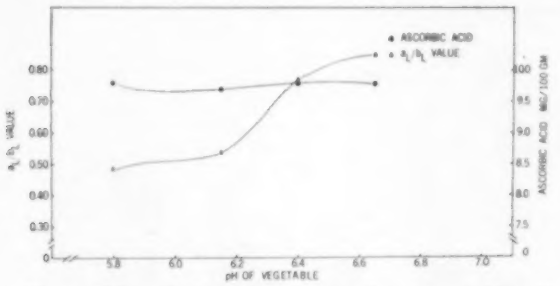

Fig. 5. Effect of pH adjustment on color and ascorbic acid in fresh green beans when cooked.

6.4 to 8.4. As with frozen green beans, buffers of pH greater than 7 resulted in only slight additional color improvement. The pH of the buffer over the range studied appeared to have no adverse effect on ascorbic acid retention.

The cooking time required for optimum texture of green beans decreased as the pH of the buffer used in the cooking liquid increased. For frozen green beans, the range was from 15 min for the control sample to 3 min for green beans cooked in water buffered at pH 8.4. For fresh green beans, it was 20 minutes for the control sample and 10 minutes for that buffered at pH 8.4.

Table 4. Effect of buffer pH on pH of cooked green beans.

	Cooking time (min)[a]	pH buffer	pH cooked vegetable
Frozen	15.0	....	5.97
	10.0	6.4	6.55
	7.0	6.6	6.65
	5.0	6.8	6.64
	5.0	7.0	6.73
	5.0	7.2	6.82
	4.5	7.6	7.04
	3.0	8.4	7.20
	1.8	8.9	7.10
Fresh	20.0	....	5.80
	17.0	6.4	6.15
	10.0	7.0	6.40
	10.0	8.0	6.65
	10.0	8.4	6.65

[a] Cooking time for optimum texture.

With both frozen and fresh green beans the effectiveness of the buffer decreased with increasing pH (Table 4). Thus, a buffer of pH 6.4 resulted in a vegetable pH not greatly different from that of the buffer. On the other hand, frozen and fresh green beans cooked in buffers of pH 8.4 had respective pH values of but 7.20 and 6.65.

Acknowledgments

The authors thank Mrs. Velma J. Chapman and Miss Patricia L. King for aid in preparation of the samples and conducting the panel flavor evaluations, and Mrs. Ada Cusick for technical assistance.

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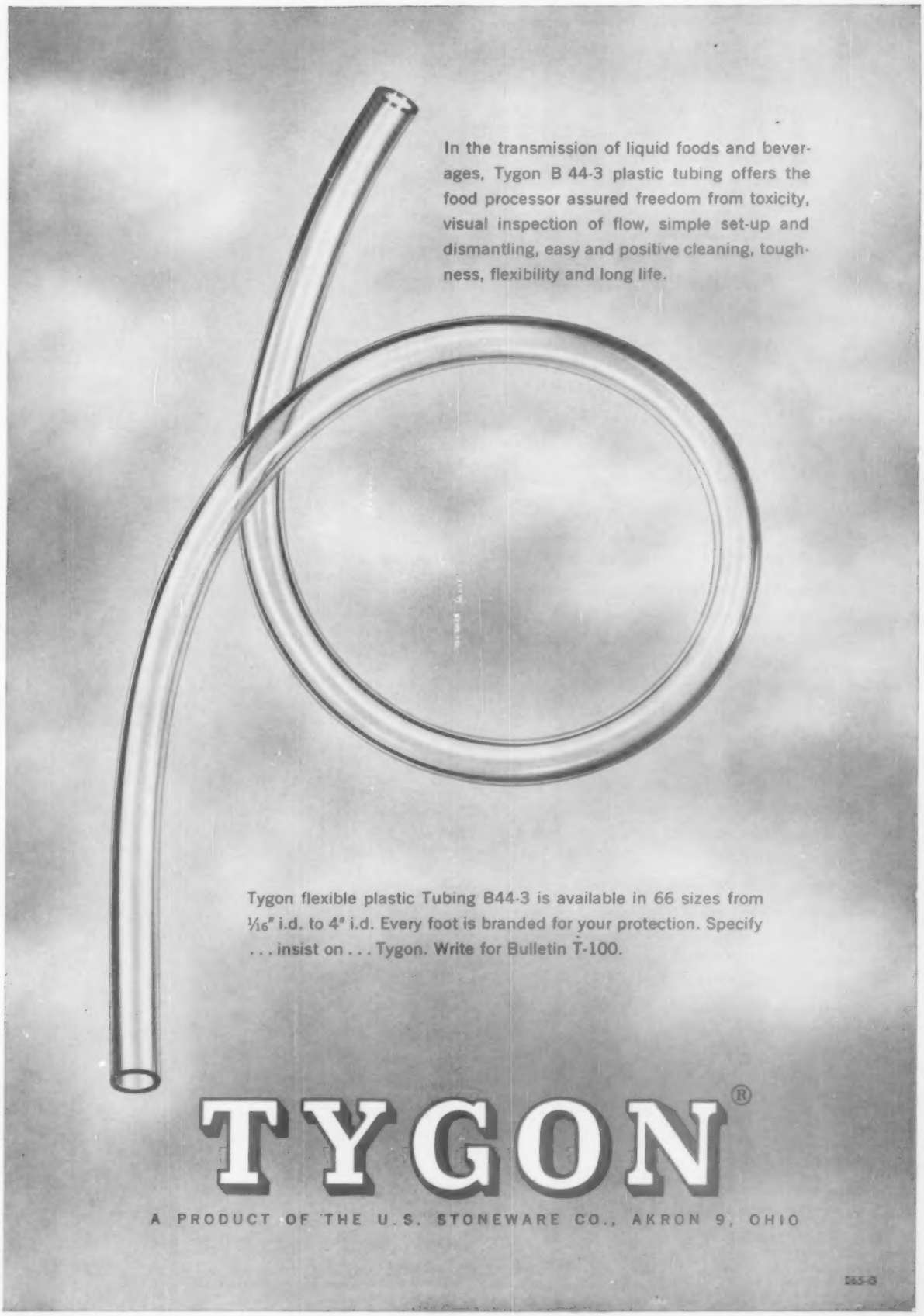
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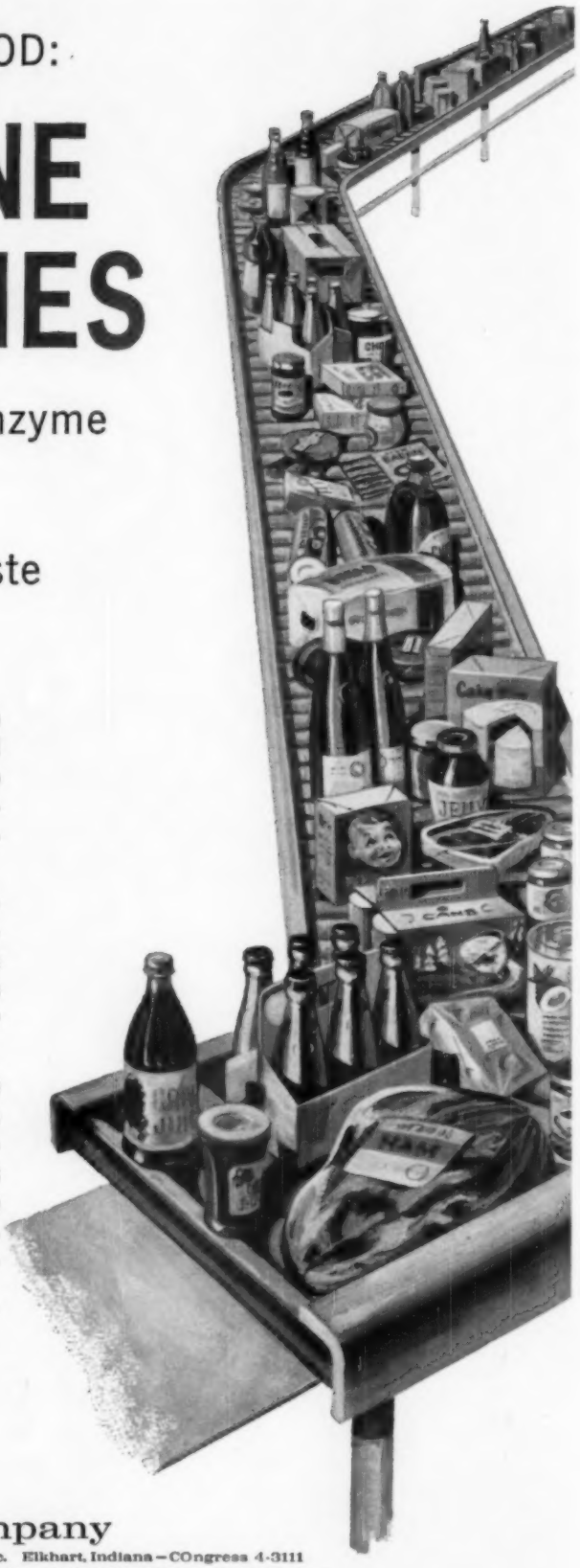

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The Effects of Smoking and Smoking Temperatures on the Shrinkage, Rancidity Development, Keeping Quality, and Palatability of Dry-Cured Hams[a]

(Manuscript received December 3, 1960)

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University of Kentucky, Lexington

SUMMARY

A total of 150 hams in two trials were dry-cured, with one group not being smoked and the other groups being heated during smoking to 80-140° F. All hams were stored 6 months at 65° F with a relative humidity of 55-60%, and then cut, observed for soundness, analyzed for salt content, and tested by a palatability panel. During aging, shrinkage was calculated and fat analyses were made for iodine numbers, peroxide numbers, and free fatty acid values. Seam fat was analyzed at 6 months in trial 2. Shrinkage increased with aging, and tended to increase with higher smoking temperatures, especially above 120° F. Iodine numbers showed no definite pattern. Peroxide numbers increased with aging, but increased faster in the unsmoked hams, followed by a smaller increase with an increase in temperature. Free fatty acids also increased with aging, but increased less as smoking temperature increased. Free fatty acid values of seam fat were lower but followed the same pattern. Hams smoked below 110°F were similar in desirable appearance and aroma and were sounder than those smoked above 110° F. Hams smoked above 110° had a lighter color, less pronounced aroma, and more undesirable green spots and deteriorated areas. Palatability scores were similar except that the higher smoked groups were more tender.

Interest has been considerable in recent years in the production of dry-cured, aged country hams in Kentucky and other southeastern states. Most early work with hams was concerned with the curing and aging processes. Methods vary considerably, causing a great variation in ham quality, as shown by Bedenbaugh and Richmond (1951), Brady *et al.* (1949), Cecil and Woodroof (1954), Fields *et al.* (1955), Hunt *et al.* (1939), Miller and Ziegler (1936), and Weir and Dunker (1953).

The methods for determining ham quality are mostly subjective. However, sampling procedures and chemical tests that objectively measure certain traits of ham quality have been described by Beasley and Carroll (1942), Blumer (1954), Hunt *et al.* (1939), and Kemp *et al.* (1957). Jensen (1949) reported that smoking cured meats has many advantages. Dunker and Hankins (1951), however, reported that only about 50% of the farmers who dry-cure meat also smoke it. These smoking procedures were carried out under various conditions of time and temperatures. Some preliminary work at the University of Kentucky showed that high-temperature smoking was detrimental to proper aging. Therefore, this project was designed to study the effect of smoking and smoking temperatures on shrinkage, rancidity, keeping quality, and palatability of dry-cured hams aged under controlled conditions of temperature and humidity.

EXPERIMENTAL PROCEDURE

Two trials were conducted. In Trial 1, 90 hams weighing 13–16 pounds were obtained from Duroc and Hampshire hogs of the Kentucky Agricultural Experiment Station. They were cured for 21 days, using 10 lb of a mixture of 7½ lb salt, 2½ lb white sugar, 2 oz potassium nitrate, and 1 oz sodium nitrite per 100 lb of ham. The mixture was rubbed uniformly over the hams on the initial, third and sixth days of curing. At the end of curing the hams were assigned to 5 groups of 18 hams each. Group 1 was wiped with a clean cloth to remove excess salt and was hung in the aging room. The other groups were washed with cold water, soaked 1 hr in a bath of lukewarm water, and hung to dry in the smokehouse. Groups 2, 3, 4, and 5 were smoked for 16–20 hr until they reached a pecan brown color and until they reached respective internal temperatures of 95, 110, 125, and 140°F. These temperatures were measured by inserting the probe of a recording thermometer into the center of the cushion of a representative ham. Heating and smoking were done in an electrically heated smoke cabinet using hardwood sawdust. The smoked hams were allowed to cool to room temperature in the smokehouse, placed in an aging room at 65°F with a relative humidity of 55–60%, and aged for 6 months.

All hams were weighed to the nearest 0.01 lb when fresh, after curing, after smoking, and at monthly intervals during aging. Fat samples weighing 0.04–0.12 lb were removed as described by Kemp *et al.* (1957) from the fresh, cured, and smoked hams and after 2, 4, and 6 months of aging. These samples were then rendered by the method of Kemp *et al.* Peroxide numbers, free fatty acid values, and iodine numbers were determined on the rendered fat samples.

The aged hams were cut and observed for color and soundness. Three adjacent ½-in. center slices were removed from 8 hams of each group. Two were used for palatability studies, and the third was analyzed for salt (NaCl). The slices were broiled in an electric oven, served to 8–12 panel members so that each member received a duplicate coded sample, and evaluated for flavor, saltiness, tenderness, and over-all satisfaction on a hedonic scale as described by Peryam and Pilgrim (1957). Results were analyzed by analysis of variance, using the method of Snedecor (1956).

Trial 2 included 6 groups of 10 hams each that were cured as in trial 1. Group 1 was not smoked, whereas groups 2, 3, 4, and 5 were heated to respective internal temperatures of 80, 90, 100, 110, and 120°F while smoking. Aging, weighing, fat sampling and rendering, analysis of outside fat, observation for color and aroma, and palatability testing were similar to trial 1 except that all hams were cut, observed, and tested. In

[a] This paper is from a project of the Kentucky Agricultural Experiment Station and is published by permission of the director.

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Table 1. Mean values for ham shrinkage.

Temp. (°F)	Cured	Smoked	Period of aging[a] (months) 1	2	3	4	5	6
Trial 1								
No smoke	2.44	.....	17.06	20.47	23.48	25.19	26.99	27.83
95	2.91	8.33	17.94	20.74	23.85	25.49	27.49	28.34
110	2.52	8.46	17.36	20.61	23.70	24.91	27.08	27.97
125	3.36	10.17	19.19	22.49	25.63	27.15	29.21	30.06
140	2.49	9.76	19.40	23.11	26.01	27.45	29.54	30.48
Trial 2								
No smoke	2.95	.....	16.10	19.96	22.56	24.61	26.40	27.86
80	2.60	6.96	15.38	19.01	21.37	23.18	24.77	26.31
90	3.66	7.92	17.02	20.79	23.51	25.65	27.46	29.05
100	3.40	7.84	16.48	19.97	22.91	25.12	26.95	28.50
110	2.61	7.17	15.44	18.84	21.26	23.28	24.91	26.42
120	3.78	9.21	18.13	22.08	24.96	27.20	29.06	30.58[b]

[a] Significant increase ($P < .01$) for each period of aging in both trials.
[b] Significant ($P < .01$) effect of smoking temperature on shrinkage in trial 2 only.

addition, seam fat was obtained from the shank portion, rendered, and analyzed for free fatty acids.

RESULTS AND DISCUSSION

Shrinkage was affected similarly in both trials (Table 1). In both trials there was a significant increase in shrinkage with each aging period ($P < .01$). Hams smoked to the higher temperatures tended to shrink more during smoking and aging. This was not significant in trial 1 but was significant ($P < .01$) in trial 2. However, the increase in shrinkage was not in direct proportion to an increase in smoking temperature. It was due mostly to additional shrinkage during smoking and only a slight increase during aging.

Iodine numbers. Iodine numbers from all groups indicated that the fat was firm. During curing and aging the iodine numbers changed but followed no definite pattern, indicating no reliable relationship between smoking temperatures and aging and iodine numbers.

Peroxide numbers. Peroxide numbers in both trials were affected by smoking, smoking temperature, aging period, and interaction between temperature and period. Table 2 summarizes the peroxide numbers. In both trials the antioxidant effect of smoke is very noticeable. In all cases the unsmoked hams showed significantly higher ($P < .01$) peroxide values at all periods after aging. Temperature also affected development of peroxide values. As smoking temperatures increased, the development of peroxides tended to decrease. This did not hold true in trial 2, however. It should be noted, however, that the temperatures were not so high in trial 2. Peroxides are intermediate products that readily break down into products such as aldehydes and ketones. As aging increased, peroxides increased significantly ($P < .01$), indicating that peroxide formation was faster than peroxide degradation.

Table 2. Mean peroxide values.

Temperature (°F)	Fresh	Cured	Smoked	Period of aging (months) 2	4	6
Trial 1[a]						
No smoke	1.96		.....	8.93	23.33	30.78
95	1.99		0.88	3.04	9.41	10.34
110	1.84		1.34	3.14	7.84	6.59
125	1.84		2.45	3.18	7.36	8.46
140	1.90		3.28	3.44	5.47	4.28
Trial 2[a]						
No smoke	2.26	2.28	.....	7.07	16.07	18.80
80	3.11	2.37	2.25	5.98	8.22	14.49
90	1.97	2.29	1.87	3.26	10.15	12.22
100	2.50	2.29	1.70	3.48	9.21	12.53
110	2.82	2.56	2.42	4.45	5.37	14.62
120	2.48	1.96	1.57	3.13	9.29	13.62

[a] Significant difference ($P < .01$)—except in trial 1, where $P < .05$ for temperature—due to temperature, periods, and interaction between temperature and periods.

Table 3. Mean free fatty acid values in outside fat.

Temperature[a] (°F)	Fresh	Cured	Smoked	Period of aging[a] (months) 2	4	6
Trial 1						
No smoke	0.25	1.28	.....	4.54	5.75	6.84
95	0.27	0.91	1.80	4.43	6.30	6.70
110	0.22	1.25	1.46	4.36	5.99	6.53
125	0.23	1.12	1.15	2.08	2.56	2.76
140	0.28	0.95	0.83	1.15	1.45	2.09
Trial 2						
No smoke	0.12	0.36	.....	3.39	6.17	9.43
80	0.11	0.43	0.53	3.16	5.90	8.65
90	0.14	0.34	0.33	3.31	5.58	8.65
100	0.13	0.35	0.50	3.04	5.11	8.21
110	0.12	0.51	0.69	2.94	5.80	7.47
120	0.11	0.33	0.63	2.08	3.32	4.90

[a] Significant difference ($P < .01$) in both trials due to aging.
[b] Significant difference ($P < .01$) in both trials due to temperature.

Free fatty acids. Free fatty acids (Table 3) showed a significant ($P < .01$) steady increase with each period of aging, a significant decrease ($P < .01$) with increase in temperature, and a significant ($P < .01$) interaction between period and temperature. Unsmoked hams developed the most free fatty acids. These results were similar in both trials. In trial 1, where the hams were heated to 125 and 140°F, there was a noticeable lack of free fatty acid development. Since free fatty acids are associated with ham flavor (Blumer, 1954), it seems that high-temperature smoking would be undesirable if flavors are to develop properly.

Free fatty acid content was determined on the seam fat of hams in trial 2. Table 4 summarizes the

Table 4. Mean free fatty acid content of seam fat from hams aged 6 months in trial 2.

Temperature (°F)	No smoke	80	90	100	110	120
FFA value	5.08	3.60	3.50	3.51	2.93	2.57[a]

[a] Significant decrease ($P < .05$) with increase in temperature.

results. The trend was the same as in the surface fat. There was a significant difference ($P < .05$) due to temperature of smoking, although the values were not as high and the differences were not as great as in the outside fat. It again shows that temperature of smoking inhibited free fatty acid development.

Salt content. The salt content of the center slices was greater in trial 1 than in trial 2. All hams were cured alike at the same temperature and for the same time. There was no humidity control in the curing room, however, so that more salt may have been lost during curing in trial 2. There is no apparent reason for a greater salt content in the 95° group, for the difference in shrinkage would not account for that much difference in salt content.

Table 5. Mean salt content of lean after 6 months of aging.

	% NaCl at various temperature (°F)					
Trial 1	No smoke	95	110	125	140	
	6.52	7.04[a]	5.72	5.72	5.70	
Trial 2	No smoke	80	90	100	110	120
	4.39	4.45	5.04	4.67	4.74	5.16

[a] Significant difference ($P < .05$) when compared with those smoked at higher temperature in trial 1 only.

Ham quality and appearance. After the hams were aged 6 months they were cut and observed for soundness, color, and aroma. Color ratings were dark red, red, light red, dark pink, pink, and light pink, respectively rated 6, 5, 4, 3, 2, and 1. Aroma ratings were typically aged, moderately aged, and slightly aged, respectively rated 3, 2, and 1. Hams smoked to temperatures above 110°F showed numerous green spots and a few deteriorated spots. These deteriorated spots are shown in Figs. 1 and 2. Table 6 shows the color and aroma scores and number of hams with spots or deteriorated places. In trial 1 there was a break in both color and aroma scores between 95°F and 110°F. In trial 2 the break came between 110 and 120°. It appears that 110°F is about the maximum internal temperature that hams should attain in smoking if they are to remain sound and develop proper color and aroma during aging.

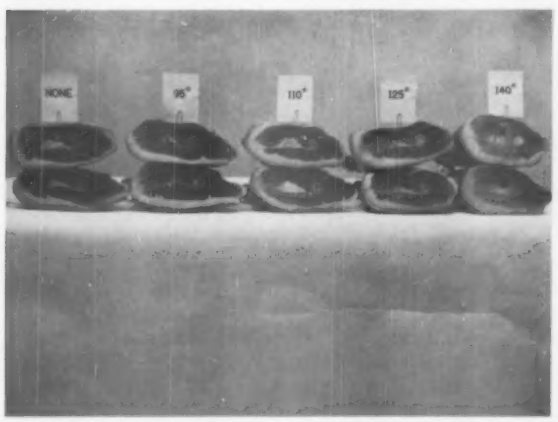

Fig. 1. Cross section of unsmoked hams and hams smoked at 95, 110, 125 and 140°F.

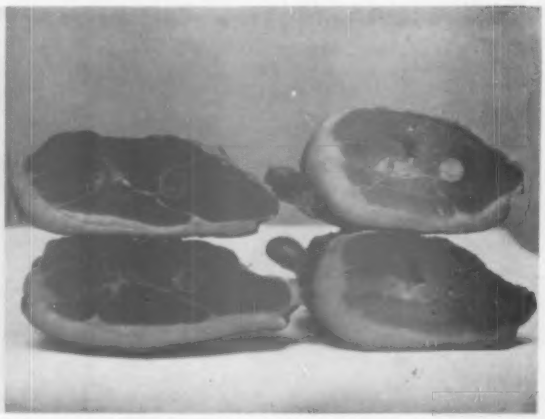

Fig. 2. Cross section of hams smoked at 95 and 140°F.

Palatability tests. Center slices from 8 hams of each group in trial 1 and all hams in trial 2 were broiled and tested by a palatability panel of 8–12 members in trial 1, and 6–8 members in trial 2. The hams were scored on a hedonic scale for flavor, saltiness, tenderness, and over-all satisfaction. Duplicate coded samples from the same muscle (either the *semitendinosus, semi-membranosus,* or *biceps femoris*) were used for each panel member. The numerical ratings used were: 9 (like extremely), 8 (like very much), 7 (like moderately), 6 (like slightly), 5 (neither like nor dislike), 4 (dislike slightly), 3 (dislike moderately), 2 (dislike very much), and 1 (dislike extremely). When hams showed green or deteriorated spots the spots were cut away before broiling so that only sound meat was tested. Table 7 gives the mean results. There were no

Table 6. Mean color and aroma scores.

	Temperature (°F)					
Trial 1	No smoke	95	110	125	140	
Color score[a]	5.5	5.8	4.8	2.7	1.4	
Aroma score[b]	2.9	2.9	2.3	1.2	1.0	
No. of hams with green or deteriorated spots	1	1	1	6	9	
	Temperature (°F)					
Trial 2	No smoke	80	90	100	110	120
Color score[a]	5.1	5.1	5.3	5.1	4.9	4.0
Aroma score[b]	3.0	2.9	2.8	3.0	2.9	2.6
No. of hams with green or deteriorated spots	1	0	0	2	2	6

[a] 1, light pink; 2, pink; 3, dark pink; 4, light red; 5, red; 6, dark red.

[b] 1, slightly aged; 2, moderately aged; 3, typically aged.

Table 7. Mean palatability scores.

	Temperature (°F)					
Trial 1	No smoke	95	110	125	140	
Flavor	6.59	6.96	6.86	6.97	6.67	
Saltiness	6.74	6.72	6.89	6.99	6.94	
Tenderness	6.38	6.69	7.21	7.64	7.86[a]	
	Temperature (°F)					
Trial 2	No smoke	80	90	100	110	120
Flavor	7.27	7.52	7.33	7.42	7.42	7.36
Saltiness	7.01	7.27	7.00	7.25	7.26	7.02
Tenderness	6.36	7.08	6.66	6.93	7.28	7.31[a]

[a] Tenderness significantly affected ($P < .01$) by temperature.

significant differences in flavor or saltiness. The hams smoked to the higher temperatures were significantly ($P < .01$) more tender, though all hams rated above 6 (like slightly).

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Gas Liquid Chromatographic Analysis of *Capsicum* Amides

SUMMARY

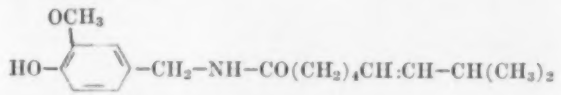
Gas chromatographic analysis of methyl esters of the fatty acids found in *Capsicum* pungent amides is capable of distinguishing between mixtures of naturally occurring and synthetic products. A small amount of an amide other than capsaicin is indicated in the natural product, with the proportion of this amide being greater in *C. annuum* than in *C. frutescens*. The results of the gas chromatographic analysis coincided with the interpretation of the infrared spectra.

THE DRIED FRUITS of two species of *Capsicum* are widely used for flavoring, and since they are generally recognized as safe by the United States Food and Drug Administration (Federal Register, 1960), they are certain to continue to be important ingredients in foods.

The pungency of *Capsicum* varies widely between varieties and between seasons, and in order to have uniform flavoring effects it is necessary to adjust formulations to take these variations into account. A number of procedures have been suggested for assaying for capsaicin, the pungent principle (Joint Committee of the Pharmaceutical Society, 1959), and the present paper is a result of an investigation to determine the cause of inconsistencies when one of these procedures was used in commercially available oleoresin *Capsicum* samples.

The method described here involves isolation of the pungent principle in a relatively pure form from the sample of oleoresin or ether extract, preparation of the methyl ester of the fatty acid portion of the capsaicin, and identification of the fatty acid or acids present by gas chromatographic analysis. The identification of capsaicin is positive, and once the absence of analogs is established, one of the procedures for determining capsaicin content may be used with accuracy.

The structure of capsaicin was shown by Nelson and Dawson (1923) to be the vanillyl amide of isodecylenic acid:



In the United States, the most common means of estimating the pungency of *Capsicum* is an organoleptic procedure introduced by Scoville (1912). This procedure will, of course, register the presence of any pungent material, but it will not differentiate capsaicin from other pungent substances, and the accuracy of the method is limited and not very reproducible between laboratories. Other methods have utilized the complexing ability of the phenolic hydroxyl group and the susceptibility of the aromatic nucleus to substitution by chromophoric substituents (Joint Committee of the Pharmaceutical Society, 1959).

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Suzuki *et al.* (1957) described a procedure utilizing the ultraviolet absorption peak of capsaicin recovered from an oleoresin by chromatographic separation. This method was used with excellent reproducibility in this laboratory on samples of *Capsicum* known to be authentic. A comprehensive review of this and similar procedures was published recently (Joint Committee of the Pharmaceutical Society, 1959). In this investigation, however, it was found that these procedures did not differentiate between capsaicin and its synthetic analogs, and it was therefore subject to reservations unless capsaicin was positively identified and the presence of other amides ruled out.

Since the amides of vanillyl amine and fatty acids such as pelargonic can be made easily and cheaply, they offer attractive adulterants for *Capsicum*. Since these amides do not have the same pungency as capsaicin, it is necessary to know of their presence if chemical and organoleptic procedures are to be related. In addition, since these amides have toxic properties (Nakajima, 1946), it is desirable to know that they are not present in *Capsicums* used as food flavor.

An earlier paper (Todd, 1958), described a simple procedure that was capable of distinguishing between the amides of capsaicin and those of saturated fatty acids. It is apparent, however, that the amides of unsaturated fatty acids could not be distinguished from capsaicin by this procedure, and this work was undertaken to provide a means of determining the specific fatty acid or acids present in the pungent amides of a *Capsicum* sample.

EXPERIMENTAL

Separation and crystallization of the pungent amide from the oleoresin or ether extract. The method follows that of Schenk (1955), which takes advantage of the very favorable separation of the pungent amides from fats, fatty acids, and pigments, using a two-phase solvent system of 80% methanol and paraffin oil. Fifty g of oleoresin and 50 g of paraffin oil are mixed, and extracted in a separatory funnel with 500 ml of 80% methanol. The methanol extract is passed through a 2.8 × 20-cm column of 1% activated charcoal (Darco KB) mixed with basic alumina, and the colorless portion of the eluate (about 400 ml) collected. This is evaporated under vacuum to about 50 ml and extracted with five 50-ml portions of ether. The combined ether extracts are dried with sodium sulfate, and the ether evaporated. (If the ether extracts are slightly colored, they should be decolorized with a pinch of charcoal before evaporation.) The residue from the ether is oily but almost pure amide, which can be used directly for preparation of esters of the fatty acid portion of the molecule or recrystallized for use in infrared spectrophotometry.

Crystallization of amides. A saturated solution of the amide in hexane is prepared by boiling the amide in hexane for several minutes (about 300 mg of amide to 100 ml of hexane). The saturated hexane is decanted from undissolved amide and added to an equal volume of boiling hexane. This is allowed to stand at room temperature in a closed container to effect crystallization, which may take 12 hours. If difficulty in crystallization is encountered, seeding with crystals of a previously prepared amide and cooling to 0° will help.

Preparation of methyl esters of the fatty acid from the amide. One-tenth to one gram of the pungent amide prepared is dissolved in 25 ml of absolute methanol, 2 ml of concentrated sulfuric acid are added, and the mixture allowed to reflux for 3 days. The solution is cooled, and 100 ml of petroleum ether added. This mixture is then neutralized rapidly with saturated sodium bicarbonate solution until effervescence stops. The petroleum ether layer is separated, dried with anhydrous sodium sulfate, and filtered, and the petroleum ether removed from the fatty esters by distillation. Complete removal is not necessary, since small amounts (50%) of petroleum fractions will not affect the gas chromatographic analysis.

Gas chromatography. The instrument used has a katharometer as detector, and a 250-cm glass column. Packing was 18% w/w Ucon 75-H-90,000 (Union Carbide) on Chromosorb "P," 35–80 mesh. Detector cell temperature was 150°, carrier gas helium 35 ml/min, at 10 lb pressure, and injection and column temperature 184°. Other packings, such as Carbowax 20,000, were equally satisfactory, as were column temperatures between 170 and 205°. Two to six μL of sample were injected, depending on the concentration of petroleum ether in the ester. Retention times were measured for the ester peaks, and the logarithm of retention time plotted against chain length for various fatty acids. Areas of peaks were determined with a planimeter.

ANALYSIS AND DISCUSSION OF RESULTS

It has been established that the logarithm of retention time (or retention volume) vs. chain length gives a straight line for methyl esters of saturated fatty acids (Keulemans, 1959). This provides a means of identifying a saturated fatty acid by retention time of its ester, if a plot is made for several known esters under the same chromatographic conditions. The location of peaks of esters made from *Capsicum* amides will likewise identify the acids present in relation to known fatty acids.

In this work, methyl esters prepared from six- to ten-carbon straight-chain saturated fatty acids were chromatographed individually and as a mixture. The ester of 10-undecenoic acid was also prepared. Fig. 1 plots retention time vs. chain length.

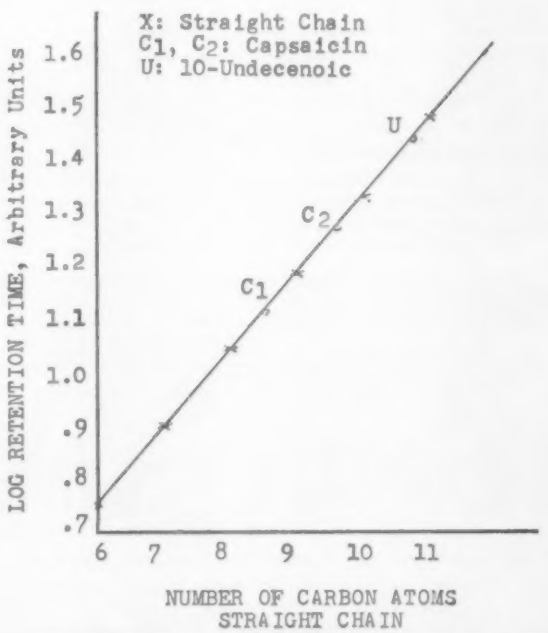

Fig. 1. Relationship of number of carbon atoms in saturated straight-chain fatty acid methyl esters to retention time, and relative retention times of capsaicin esters and 10-undecenoic ester.

Synthetic capsaicin analogs were made by condensing vanillyl amine and the acid chlorides of pelargonic and 10-undecenoic acids. The methyl esters prepared from these amides had the same retention times as the esters prepared directly from the respective acids.

A chromatogram of the methyl esters prepared from a sample of crystalline capsaicin showed two peaks, designated c_1 and c_2. c_1 decreases relative to c_2 with recrystallization, indicating that the main peak is that due to isodecylenic acid.

The retention times of the two capsaicin ester peaks are shown in the plot in Fig. 1. c_1 may be a fatty acid homologous to isodecylenic, but one carbon shorter. No attempts were made to identify this ester further at this time.

It should also be mentioned that there was an indication of an ester with a retention time between c_1 and c_2 in the ester prepared from uncrystallized capsaicin, but that this peak was so small as to be indefinite.

To determine the applicability of this method of analysis to various types of *Capsicums*, a number of varieties were extracted and analyzed. All samples gave two peaks, with the relative amount of c_1 being greater in the varieties classified under the species *annuum*. These results are summarized in Table 1.

Table 1. Effect of Capsicum species on proportion of c_1 fatty acid to c_2 fatty acid, and the location of the ethylenic stretching mode in the infrared spectra of their capsaicin.

	% total area of esters in c_1 peak	Location of stretching mode peak (cm^{-1})
Capsicum frutescens		
Mombassa	2.2	970
Sudanese	4.0	970
Capsicum annuum		
Hungarian	7.4	970
Carolina long	7.8	970

The method was also applied to samples of oleoresin *Capsicum* obtained through commercial channels. The results are in Table 2. Sample 1 is the same material in which pelargonamide was identified in a previous paper (Todd, 1958), and its presence is again confirmed.

Table 2. Fatty acid esters observed in commercial oleoresins of Capsicum.

Sample No.	c_1 and c_2 (*Capsicum* peaks)	Normal saturated fatty acids with carbon atoms numbering			
		7	8	9	10
1				X	
2	X				
3	X			X	
4	X	X	X	X	X
5	X				
6				X	

Attempts were made to utilize infrared spectra to corroborate the findings further. Capsaicin in CCl_4 was found to have a peak at 970 cm^{-1}, and the vanillyl amide of 10-undecenoic acid was found to have a peak at 913 cm^{-1} on a Beckman IR-5. The latter peak is obtained from 10-undecenoic acid itself, and has been interpreted as the olefinic C-H stretching mode. No peak in this region is observed with the pelargonic acid derivative. The amide must be recrystallized before infrared spectra are run, since the crude amide used in preparation of the ester may contain interfering substances.

In all samples in which the chromatographic analysis showed capsaicin to be absent, the infrared spectrum of the amide did not show a peak at 970 cm^{-1}. In those samples containing capsaicin alone or in combination with other amides, the infrared spectrum showed this peak.

Since the peak at 970 cm^{-1} appears to be unique to capsaicin, it is useful for determining the presence of capsaicin. However, infrared will not portray the presence of a mixture of fatty acid amides as will gas chromatography, and for this reason the gas chromatographic analysis provides the most information about the composition of a sample.

Acknowledgment

The assistance of R. W. Clement in running and interpreting the infrared spectra is appreciated.

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October 17, 1960.

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(Continued from Page 31 preceding Technical Section)

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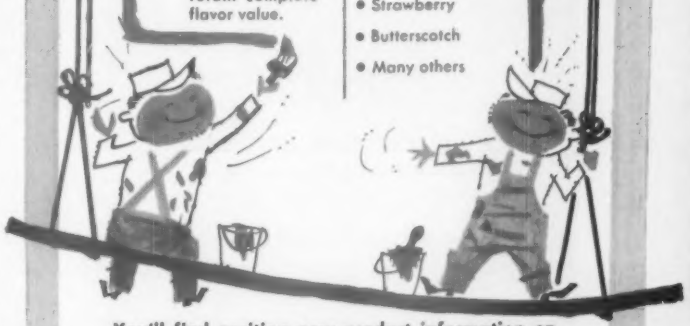

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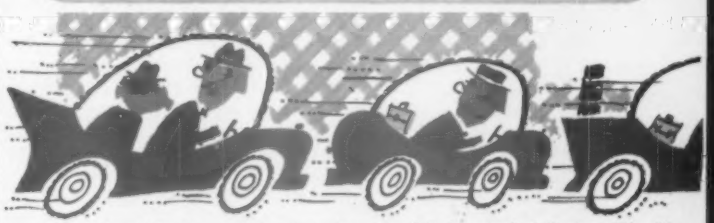

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Although the London Congress is being organized by the host country, speakers and participants will come from many countries. Their papers will cover these general fields of interest:

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- Quality, Analysis, and Composition of Foods
- Manufacture and Distribution of Foods

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CONGRESS SYMPOSIA

Education and training of food scientists.

Changing patterns of consumption of food (national and international).

Effects of modern agricultural practices on foods.

SECTIONS

SECTION A. Chemical and Physical Aspects of Foods

Proteins
- Chemistry of muscle and connective tissue; related enzyme systems.
- Proteins of eggs and milk; related enzyme systems.
- Vegetable proteins.

Carbohydrates
- Pectin, hemicelluloses, and plant gums; microstructure of cell walls.
- Cellulose, starch, glycogen; related enzyme systems.
- Sugars, amino sugars, and glycosides: browning reactions.

Fats
- Composition and properties of lipids, lipoproteins, carotenoids, and steroids.
- Deteriorative changes in lipids; rancidity, antioxidants, lipases.

Phenolic and Related Substances
- Natural pigments.
- Tannins and lignins.

Biochemistry of Ripening
- Fruits and vegetables.
- Seeds.

Food Additives and Contaminants
- Preservatives and coloring matters.
- Toxicology.

Radiation Preservation
- Chemical effects: flavor, odor, appearance.
- Effects on enzyme systems and nutritive value.

Physics and Biophysics
- Biophysical techniques; e.g., electrophoresis, infrared spectra, light scattering, viscosity.
- Physics of drying, freezing, heat transfer, and evaporation.
- Physical properties of foods: rheology, emulsions, surface activity, osmosis.

SECTION B. Biological and Microbiological Aspects of Foods

Effect of Conditions of Husbandry and Cultivation on Quality of Foods
- Fruit.
- Vegetables.
- Meat and fish.
- Oil seeds.

Physiology of Maturation, Transport, and Storage of Foods
- Fruit.
- Vegetables.
- Meat and fish.
- Oil seeds.

Food Preservation
- Preservation by traditional methods (e.g., sugar, salt).
- Inhibitory substances: natural and other inhibitory substances.
- Physical methods.

Microbial Ecology of Sound and Spoiled Foods
- Natural foods.
- Matured and stored foods (including curing, pickling, etc.)

Hygiene
- Microbiological assessments of quality: fresh foods, frozen foods, processed foods.
- Food processing plant.
- Pest control.
- Waste-product recovery and utilization.

Industrial Fermentations
- Accessory food substances.
- Other food substances and newer methods.

Laboratory Techniques
- Isolation and identification of microorganisms.

SECTION C. Quality, Analysis, and Composition of Foods

Specialized Analytical Techniques
- Routine control, speed of operation, and microbiological assays.

Detection and Determination of Minor Constituents
- Nutritional significance: vitamins, amino acids.
- Technological additives: antibiotics, antioxidants, colors, emulsifiers.
- Adventitious contaminants: residues from agricultural pesticide treatment, residues from hormone implantation.

Application of Automation to Analysis and Quality Control
- A review.

Subjective and Objective Methods of Quality Assessment
- Use of consumer surveys.
- Training and use of panels.
- Identification of flavor constituents.
- Measurement of texture.

Establishment and Operation of Quality Control
- A review.

Changes in Quality Due to Processing
- Heat.
- Dehydration.
- Freezing.
- Irradiation.

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Changes in Quality Due to Storage
A review.

Composition of Raw Materials and Manufactured Goods
A review.

Variation of Raw Materials in Relation to Food Processing
Effects of feeding and hormone treatment on animal products.
Effects of cultural treatment on vegetable products.

SECTION D. Manufacture and Distribution of Foods

Dehydration
Effects of selection of raw materials.
Pre-treatments: fruits and vegetables, meats.
Drying techniques: spray, drum, film, vacuum, freeze.
Storage problems: gas and vacuum packing, antioxidants, in-pack desiccants, moisture content.

Freezing
Effects of selection of raw materials.
Pre-treatments with special reference to blanching.
Quick-freezing techniques: air blast, immersion, plate bulk freezing.

Canning
Effects of selection of raw materials.
Pre-treatments.
Canning techniques: continuous cookers, hydrostatic cookers, end-over-end canning, hot-air sterilization, other heat-transfer media.
Can design, including corrosion and staining.
High-speed filling and closing machinery.
Post-processing infection.

Pasteurization and Sterilization
Gamma rays and fast electrons.
Ultra-high temperature.
High-frequency.
Infrared.
Micro-waves.

Packaging
Materials.
Machinery.
Special processes, e.g., vacuum and gas.

Distribution
Cold-storage design.
Automatic vending.
Fresh and frozen products.

General Problems in Food Factories
Effluents.
Hygiene.
Instrumentation and control.

Agricultural Processes
Application of chemicals to crops.
Mechanical harvesting.
Mechanical grading.

Manufacturing Processes
Cereals.
Bread and flour goods.
Starch, sugar, and chocolate goods.
Processed fats.
Meat and fish products.
Fruit products.
Vegetable products.

During the Congress, arrangements have been made to visit food factories, universities, and government and industrial laboratories. Social events will include a reception and a banquet.

Further information on the Congress may be obtained from the Honorary Secretary, First International Congress of Food Science and Technology, 14 Belgrave Square, London, S.W.1.

FUTURE MEETINGS FOR FOOD TECHNOLOGISTS

1961	
May 15-18	Fourth International Congress on Canned Foods, Berlin, Germany (for information address M. Henri Cheftel, 71, Avenue Edouard Vaillant, Billancourt (Seine), France.
May 17-21	British National Association of Wholesale Distributors of Frozen Foods, Yorks and Old Swan Hotels, Harrogate
May 24-26	Canadian Institute of Food Technology, Queen Elizabeth Hotel, Montreal
May 30	Nutrition Society of Canada, Ontario Agricultural College, Guelph, Ontario
June 5-9	Ninth National Plastics Exposition, New York Coliseum
June 13-16	3rd Biennial International Gas Chromatography Symposium, Kellogg Center, Michigan State University
June 15-17	Shrimp Association of the Americas, Mexico City
June 18-23	Association of Food & Drug Officials of the United States, Mayflower Hotel, Washington, D. C.
June 27-30	American Home Economics Association, Auditorium, Cleveland, Ohio
June 19-23	42nd Annual Meeting, Pac. Div. Amer. Assn. Advancement of Science, Univ. of Calif., Davis.
July 10-14	National Food Distributors Association, New York City
Sept. 15-19	American Meat Institute, Palmer House, Chicago, Illinois
Sept. 19-22	Australian Conference on Food Technology, C.S.I.R.O., Homebush, Australia
Sept. 19-27	International Conference on Fish in Nutrition, Washington, D. C.
Sept. 28-30	American Hotel Association, Washington, D. C.
Oct. 7-12	Baking Industry Exposition, Convention Hall, Atlantic City
Oct. 15-18	National Association of Food Chains, Palmer House, Chicago, Illinois
Oct. 18-20	National Packaging Forum of Packaging Institute, Biltmore Hotel, New York City
Oct. 24-27	American Dietetic Association, Jefferson Hotel, St. Louis
Nov. 5-8	National Frozen Food Association, Bal Harbour and Americana Hotels, Miami Beach
Nov. 6-10	National Hotel Exposition, Coliseum, New York City
Nov. 12-15	Air Conditioning and Refrigeration Institute, Homestead Hotel, Hot Springs
Nov. 15	British National Association of Wholesale Distributors of Frozen Foods—Annual Lunch, Trocadero, London
Nov. 27-Dec. 1	28th Exposition of Chemical Industries, New York Coliseum
1962	
Feb. 9-11	Institute of American Poultry Industries' 33rd Annual Fact Finding Conference; Municipal Auditorium, Kansas City, Missouri
June 10-14	**Twenty-second Annual Meeting of the Institute of Food Technologists, Fontainebleau Hotel, Eden Roc Hotel, Miami Beach, Florida**
1963	
May 19-23	**Twenty-third Annual Meeting of the Institute of Food Technologists, Pantlind Hotel, Civic Auditorium, Grand Rapids, Michigan**

An open invitation is extended to readers of FOOD TECHNOLOGY to send in to the Editorial Office, P. O. Box 164, Davis, California, notices of annual or national meetings of interest to food technologists.

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International Food Technology

In September, 1960, Dr. Siegfried S. Lichtblau assumed Directorship of the Food Industries Advisory Service (FIAS), 22 Jerusalem Boulevard, Jaffa, Israel. The Service is a consulting and information center which, according to Dr. Lichtblau in a recent letter, is supposed to develop into a central food institution in Israel.

Since assuming the post, several well-known scientists have visited the organization including Dr. Donald K. Tressler, who remained for ten weeks. Dr. Berton S. Clark is currently serving the FIAS in an advisory capacity with respect to the metallurgical side of canning. He will remain there until the middle of July.

According to Dr. Lichtblau, the FIAS is composed of 65 members, mostly industries, but also institutions like the new Sheraton Hotel in Tel Aviv. The organization maintains a library, acts as a consultant, and is in contact with numerous sources of information about the food industry, both within and without Israel.

International Food Technology—

The sixth symposium on materials of foreign origin in food was conducted in mid-October, 1960, at the National Center for Agronomic Research, Madrid, Spain.

The seven-day conference was divided into several sections, with discussion of processing methods employing bacteria, molds, yeasts, and enzymes, and the uses and residues of antibiotics in animals and poultry. A large section was devoted to the relation of safety, purity, nutritional value and flavor to foods produced by fermentation.

Some of the participants were: A. Barret, Versailles, France; J. M. Rodriquez Bordolia, Seville, Spain; Edouard Brochu, Oka-La Trappe, Canada; H. Fink, Cologne, Federal German Republic; B. P. Eddy, Cambridge, England; Claudio Antoniani, Milan, Italy; J. Hollo, Budapest, Hungary; G. Mackinney, Berkeley, California; and Tetuo Tomiyama, Fukuoka, Japan.

Nominations for Packaging Awards Will Close May 15

Nominations for two annual awards given by the Packaging Institute will close May 15. Candidates need not be Institute members.

The Institute Corporate Award is given to the company which has made an outstanding advance in applied packaging technology.

The Professional Award is given to the individual who has made an outstanding contribution to packaging technology.

All nominations must be accompanied by ten copies of the original entry form which may be obtained from the Packaging Institute, 342 Madison Ave., New York 17, N. Y.

Translations of Three Soviet Fisheries Reports Are Issued

A critical examination by Soviet scientists of theories on the pink salmon's biennial life cycle, and a report of investigations into the biological factors responsible for fluctuations in the supply of this fish, are given in a Russian bulletin now available in English from the Office of Technical Services, Business and Defense Services Administration, U. S. Department of Commerce, Washington 25, D. C.

Also available from the same source are translations of two other papers, one on methods for increasing the supply of Amur pink salmon, and the other on processed smoked fish.

The three reports are:

Age of Pink Salmon and the Pattern of their Fluctuations in Abundance, 60-21150. Translated from USSR Bulletin of the Pacific Research Institute for Fisheries and Oceanography, Vol. 41, 1954. 114 pages, price $1.25.

The State of Stocks and Means for Increasing the Number of Amur Pink Salmon, 60-21144. Translated from Proceedings of the Conference on Problems of Pink Salmon Economy in the Far East, No. 4, 1953, Moscow. 35 pages, price 50 cents.

Histological and Histochemical Investigation of Tissues of Smoked Fish, 60-21132. Translated from Fish Economics, Vol. 35, No. 2, 1959, Moscow. 8 pages, price 50 cents.

REGIONAL NEWS

CHICAGO

Federal laws and regulations are a necessary part of the food industry, Dr. Imri Hutchings, president of the Institute of Food Technologists, told the Chicago Section of IFT during its February meeting.

He, personally, is a strong supporter of the regulations of the Food and Drug Administration, Dr. Hutchings said, and his company, the H. J. Heinz Company, has been a willing cooperator with the federal office since its inception.

However, there has been a change in the type of cooperation from the Food and Drug Administration. This change dates from the adoption of the Public Law 929, the Food Additives Amendment. "There seems to be little need for this change in direction," Dr. Hutchings said, "and I fear this is only the beginning of more stringent controls."

Dr. Hutchings reported that the former secretary of the Department of Health, Education and Welfare, Arthur Flemming, said that the Food and Drug Administration is responsible for maximum protection to consumers. Dr. Hutchings emphasized the word "maximum," in Flemming's declaration, noting that it was given no relation to economic feasibility.

The problem is compounded by the attitude of consumers. "Maximum" protection sounds good to them. Dr. Hutchings quoted from a letter which appeared

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in a "letters to the editor" column of a trade paper, a housewife was "thankful that Mr. Flemming had the courage to stand up against the big food manufacturers who try to force their poisonous products on an unsuspecting public." This woman, Dr. Hutchings said, is not alone.

For example, at a Federal Law Institute banquet, Congressman John E. Fogarty said that his committee had given the FDA several million dollars more than it had requested. He turned to a FDA representative on the speakers' platform with him and said, "you can have as much money as you want, to keep the food industry from poisoning the country."

The government, supported by popular approval, is prepared to move in and put the food industry under strict and stringent control, Dr. Hutchings believes. Statements made by the FDA on the standards the food industry should meet, clearly imply a supposed inadequacy of the food industry and infer that the government will find it necessary to move in and take charge.

In order to forestall such an action Dr. Hutchings advised his fellow technologists to face up to their responsibilities to the consumer to produce for quality rather than just profit. "We as food technologists have a responsibility to keep our management in perspective," he said. Some technologists did not have enough guts to do this, he regretted.

Further, he said, we must recognize that our food technologists must be well trained. "We don't want food technology mills to grind out second-rate graduates." As manager of food research for the H. J. Heinz Company, Dr. Hutchings said that he had great difficulty in finding individuals properly trained for technology. We are not training well-rounded technologists at the present time, he said. He felt that higher qualifications were necessary.

Better liaison is necessary between the food industry and federal agencies. The recent fiasco with FDA Red No. 1, which was recently and publicly taken off the approved list for use in foods such as maraschino cherries, has led Dr. Hutchings to believe that the action was taken out of ignorance. The FDA needs help in getting more information. He said that a group of technologists will serve as consultants to FDA.

Finally, each technologist should take an active part in general public education. He should offer to sell food technology at every opportunity.

—Dennis Murphy

Book Review

METHODS OF BIOCHEMICAL ANALYSIS. Vol 8. Edited by David Glick, Professor of Physiological Chemistry, University of Minnesota. Interscience Publishers, Inc., 250 Fifth Avenue, New York 1, N. Y. 1960. IX + 400 pp. Price $10.00.

This publication is a continuation of an annual series of volumes, started in 1954, to help the research worker keep abreast of the latest techniques and methods in specialized areas of biochemistry.

The first chapter (A. T. James) is concerned with qualitative and quantitative determination of the fatty acids by gas-liquid chromatography. The first part of this chapter, on the apparatus and techniques used in gas-liquid chromatography, should prove of value to all those interested in one of the newest and most sensitive techniques of separation and analysis. The second part deals with preparation of the sample of fatty acids, separation, and analysis of the data. The author has included a great deal of important unpublished data from his own laboratory, and a large number of "don'ts" and "cautions." It is an excellent chapter, in both content and clarity, and should be invaluable to the biochemist working in the area of lipid composition and biosynthesis.

The second chapter (A. M. Chase) is concerned with measurement of luciferin and luciferase in the three bioluminescent systems from which these compounds have been partially purified: luminous bacteria, the firefly, and the ostracod crustacean *Cypidina hilgendorfii*. Much of this chapter is devoted to details of the apparatus used for measuring light emission from these systems. Details are included for purification and assay of these systems, but the author complicates knowledge of the methods of measuring luciferin and luciferase more than he brings order out of chaos. One is left with the feeling that one must consult the original papers after all, in order to be able to work with these systems. The section on application of bioluminescent systems to measurement of adenosine triphosphate and coenzyme A, the detection of traces of oxygen, and other possible uses should be of general interest.

The third chapter (P. Talalay), on enzymic analysis of steroid hormones, reviews the preparation and use of hydroxysteroid dehydrogenases for the specific and sensitive microdetermination of the steroid hormones, their precursors, and degradation products. The author certainly demonstrates why the enzymatic methods may largely replace the relatively insensitive and unspecific chemical methods now employed for assay of this group of compounds. He points out the need for the significance of the discovery and purification of other enzymes (in addition to the alpha and beta hydroxysteroid dehydrogenases described here) capable of using other sterols (or other positions on the sterol) as substrates. This chapter is recommended to those working in the area of sterols, particularly those in the medical profession or in the area of sterol biosynthesis, and to those interested in the area of practical enzyme applications.

The fourth chapter (M. E. Scott) is concerned with application of aliphatic ammonium salts in the assay of the acidic polysaccharides such as chondroitin sulfate, heparin, carrageenin, and pectin from tissues. This chapter deals largely with the physical and physicochemical aspects of the problem. Emphasis is on principles, general procedures, and typical examples rather than on the quirks and peculiarities of isolated systems. This chapter will be invaluable for

those working in the chemical and biochemical aspects of acidic polysaccharides found in all plant and animal tissues, and is recommended to those interested in the removal of these materials from certain foods.

The fifth chapter (M. W. Whitside and F. Zilliken) is on isolation and determination of neuraminic (sialic) acids. Methods of analysis of sialic acids are particularly appropriate at this time, when a great deal of attention is being given to these aminocarboxyoctuloses, which are widely distributed throughout the animal kingdom, especially as components of the mucoproteins of blood, mucin, milk, egg white, etc., and have been found in certain bacteria. The authors give quite detailed procedures for the isolation of *N*-acetylneuraminic acid (from human milk, bovine submaxillary mucin, and *Escherichia coli*) and methoxyneuraminic acid (from urinary mucoproteins and mucolipids) and their identification and characterization by X-ray diffraction, infrared spectroscopy, and chromatography, and quantitative determination by colorimetric procedures. The authors have done an excellent job of presenting, in detail, the methods of analysis in only 21 pages.

The sixth chapter (S. Schwartz, M. H. Berg, I. Bossenmaier, and H. Dinsmore) is concerned with the determination of porphyins in biological material. This chapter gives major emphasis to a consideration of the general principles derived from the physicochemical properties of porphyins. Some of the more important specific analytical procedures are described, plus a consideration of alternative possibilities, limitations, and areas for future development. This chapter deals mainly with the porphyins of urine, feces, and erythrocytes, but not with hemoglobin and myoglobin. The coverage is comprehensive (approximately one-fifth of the volume), and the large collection of references should be invaluable. This chapter is intended primarily for those interested in the medical aspects of porphyins, but will be of value to other biochemists in this area.

The seventh chapter (A. B. Lerner and M. R. Wright) describes techniques in which isolated pieces of frog skin are used for quantitative determination of melanocyte-darkening and -lightening agents. These techniques are surprisingly sensitive (10^{-12} molar MSH can be detected), but suffer from the same disadvantages of all bioassays. This brief chapter will be of interest to the worker starting in this field, but will be of little use to the established worker.

The eighth chapter (C. Kitzinger and T. H. Benzinger) is concerned with the principle and method of heatburst microcalorimetry of energy changes. It deals with measurement of the changes in energy of a reacting biochemical system rather than with the changes in matter. A detailed account is given of the construction of a microcalorimeter that avoids elaborate temperature controls, constant-temperature baths, and complex electronic equipment. The instrument detects extremely small changes in heat quantities with great speed, as indicated by the applications in which it was used. The authors have applied this technique to measuring changes in enthalpy of asparagine and adenosine triphosphate hydrolysis and an antigen-antibody reaction, and changes in enthalpy, free energy, and entropy of the reactions of fumarate to malate conversion and glutamine hydrolysis. The method is so sensitive that it has been used in determining the purity of glutaminase. This chapter and method should be given careful consideration by all biochemists interested in measuring energy changes in reaction systems.

This volume is a valuable addition to this series and will find a great deal of use by biochemists working in the areas described within the book. Chapter 8, and chapters 1 and 4 to some extent, help to make this volume of general interest.

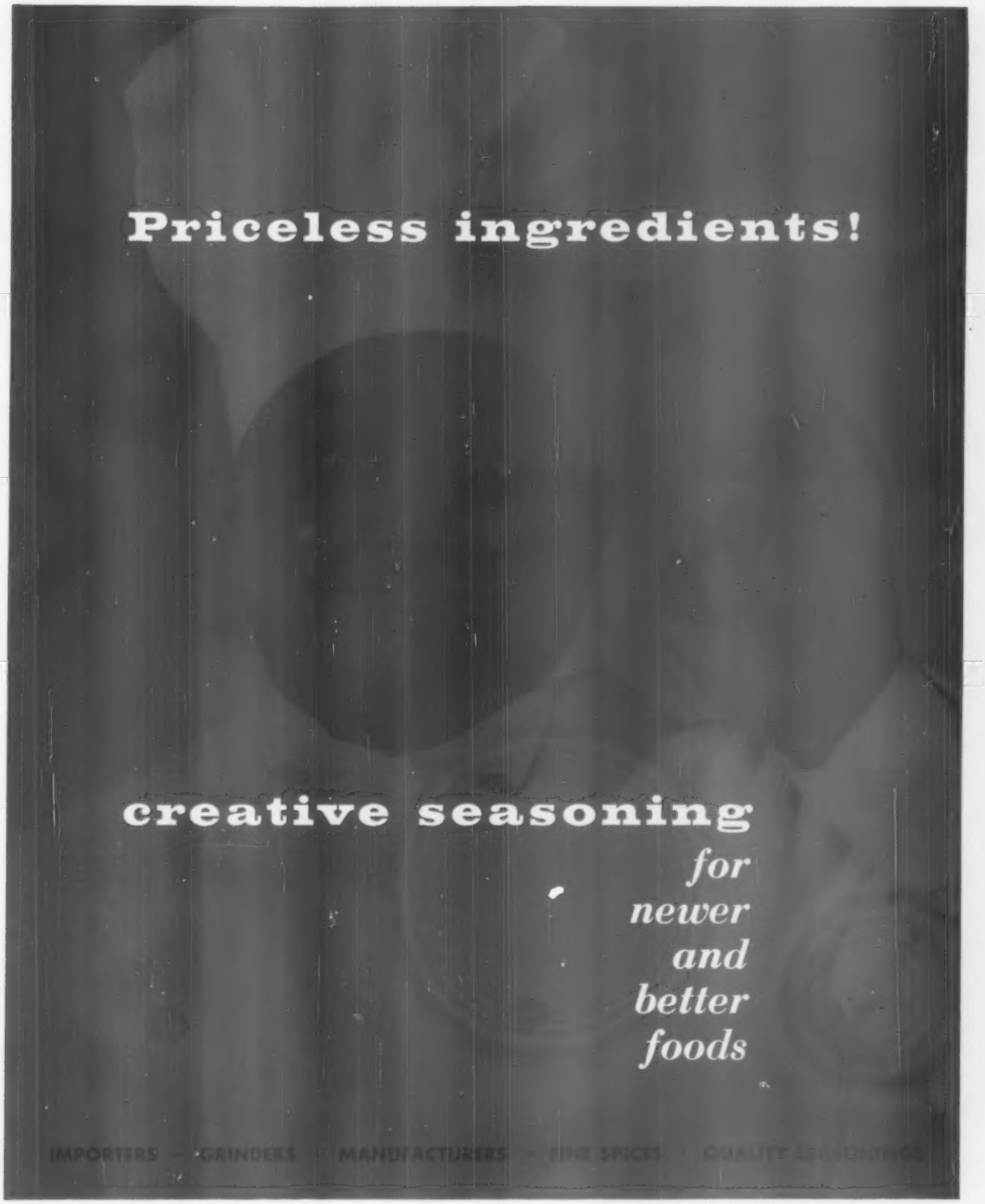
JOHN R. WHITAKER

MICROSCOPIC–ANALYTICAL METHODS IN FOOD AND DRUG CONTROL. Harris, K. L. (Editor). Food and Drug Technical Bulletin No. 1. U. S. Department of Health, Education, and Welfare. 1960. 255 pages. $2.00.

This bulletin replaces an earlier publication, *Microanalysis of Food and Drug Products*, which was Food and Drug Circular No. 1. The new publication is a cooperative venture by members of the Division of Microbiology of the Food and Drug Administration, U. S. Department of Health, Education, and Welfare. Essentially, the purpose of the authors is to provide a background in microscopy which will be useful to analysts in regulatory and control activities. However, the bulletin is envisioned as being more than a manual: it aims "to show the scope of this microscopic analytical work, the approach used, the application of such techniques to industry and regulatory problems, and the ways in which results can be interpreted." Thus, it is hoped that the publication will also serve the needs of administrators, attorneys, and jurists—with the note, however, that "detailed application requires experience with reference and authentic materials."

The first two chapters are directed to the general reader who wants to comprehend the relationship between sanitation and contamination and the rôle of inspection procedures in this relationship. In this section there is also provided a legal background and a rationale for the inspection program of the Food and Drug Administration in the United States. Although Chapter 3 leaves the general background in order to take up specific problems of sample isolation and inspection techniques, the material given here could well be read with profit by the non-specialist. The presentation is highly readable.

Chapter 4 gives the principles of microscope construction, together with helpful suggestions for the use of microscopic equipment. Particular attention is paid to photomicrography as applied to exhibit preparation. Chapter 5 (20 pages) is devoted to the fungi associated with food decomposition, Chapter 6 (83 pages) to insects in foods, Chapter 7 (10 pages) to parasitic Protozoa and helminths, Chapter 8 (21 pages) to rodent and other animal filth, Chapter 9 (11



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pages) to plant histology, and Chapter 10 (30 pages) to crystallographic study with polarized light. The final chapter (Chapter 11) is extremely short, dealing with examination of drug tablets and capsules.

In essence, the broad aims of the bulletin have been served well. Within the limitation of the number of pages available and in cognizance of the probable background of the average analyst, the authors have accomplished a remarkably well-documented job. This reviewer was not completely satisfied with the degree of emphasis on each topic, but perhaps that has been governed by considerations other than the one of uniform treatment. Within the purview of the principal goal, that of training an analyst, the balance of emphasis here may well be the most useful one.

Several topics have been completely omitted. A future edition should include a discussion of microchemical test methods to distinguish between food tissues and the contaminants. These tests have been particularly well developed in recent years and constitute a powerful adjunct to the morphological methods described here. It is regrettable that no material on animal tissues has been presented and that the illustrations for plant histology and organography are so limited. Professor E. M. Mrak has complained about the lack of histological training for food technology students: "They see a germinating seed of tomato and call it a worm!"

Against these minor criticisms, there are many major positive aspects in this work. The material is well-organized, and the text is abundantly illustrated with striking photographs and photomicrographs. (Although many of the line-drawings are excellent, some seem to be rather uneven.) Each chapter has a bibliography which is quite useful, even though a few references are not up-to-date. However, the inclusion of bibliographic material is a marked improvement from the earlier, unrefereneed publication. Particular note should be paid the section on animal hairs in Chapter 8. This is most original and useful information which was not presented at all earlier.

The work is well-written, and the publication is a visual delight. Undoubtedly, this bulletin has cost far more to produce than the price which is being asked. For the extensive use that it will receive from its principal audience of analysts and administrative personnel, the publication perhaps should have been bound in hard, rather than soft, covers.

CLARENCE STERLING

FOOD FOR AMERICA'S FUTURE, 1960. Ethyl Corporation, McGraw-Hill Book Co., Inc., Publishers, 330 West 42nd Street, New York 36, New York, 167 pp.

This timely little book is a splendid non-technical description of the means presently available or just over the horizon for feeding the U.S.A.'s 245 million people expected in 1975. It is divided into 12 chapters, each by a different author. In a sense, each chapter is a separate essay on a question something like: "Will there be enough food by 1975 to maintain present levels of consumption?" The style of writing indicates, however, that there was a common editor. Therefore, although there is some repetition, the book generally flows smoothly and reads easily.

Firman E. Bear, Editor-in-Chief of *Soil Science*, points out that there are presently about 11 acres of land in the United States for each of the 180 million people; by 1975 the area per person will not exceed 9 acres; and by the year 2000 will be down to 6 acres. More than half of our land is not suitable for growing harvested crops. He also indicates that we are presently using about 350 million acres of cropland. He believes that this area could be doubled through irrigation, drainage, chemical treatment, or changed land use; and once the whole secret of plants' ability to manufacture sugar is learned, assuming adequate energy sources, this country ought to be able to produce enough food to feed a billion people and feed them well.

Byron T. Shaw, Administrator of U.S.D.A.'s Agricultural Research Service, emphasizes the continued need for research to provide food for the future. Farmers must more than double the record-setting annual increases in output that have occurred since World War II to meet the needs of 1975—an increase of 35–40% during the next 15 years. Rogers and Barton, Agricultural Economists of the U. S. Department of Agriculture, have indicated that farm output could be expanded 35% by 1975 through application of presently known technology. (Rogers, Robert O., and Glen T. Barton. *Our Production Potential, 1975*, Agr. Info. Bulletin No. 233, U. S. Department of Agriculture, 1960.)

Thomas A. Boyd, a close associate of Charles F. Kettering, writes about Mr. Kettering's study of plant growth. This chapter contains much of Kettering's optimistic philosophy. Despite the gap between present knowledge and application, few social scientists, if any, including Rogers and Barton, believe that research on physical or biological problems of agriculture should be stopped until the gap is closed. One quotation from Kettering emphasizes his point-of-view: "Research is something which, if you don't undertake until you have to, it is too late. It is just as though you waited to see the smoke rolling out of a building before calling a fire insurance agent."

Richard C. McCurdy, President of Shell Chemical, discusses the need to control insects, diseases, and weeds to meet the needs of the future. He appears to forget about the low elasticity of demand for foods in discussing present dollar losses from insects, but believes that with good research programs we are likely to develop new insecticidal weapons faster than insect pests develop resistance against old ones.

Kenneth S. Adams, Chairman of the Board of Phillips Petroleum, believes we must maintain the productive power of our land through cropping practices. He uses a rather odd method of justifying greater forage utilization—the low recovery of food value from grains by meat animals. He does not show that greater forage use would result in greater efficiency of resource use. He does an excellent job of

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justifying his optimism about the availability of supplies of fertilizer and other soil-improving substances, industry's ability to meet the challenge of supplying the plant-food needs of tomorrow's people, and agriculture's ability to close the narrowing gap between knowledge and application.

Robert S. Stevenson, President of Allis-Chalmers, anticipates an additional 2¼ million tractors—50% more than presently—on U. S. farms by 1975. He also expects the trend toward more efficient tractors to continue. Dealers will become application engineers, and industry will be able to supply the equipment for power farming.

Robert E. Wilson, Chairman of the Board (Retired) of Standard Oil of Indiana, sees no shortage of liquid fuels. Among the major new sources are the huge deposits of shale oil from which gasoline can presently be made at a cost of about 30% more than from petroleum. The major question to him is not whether farmers will be able to obtain oils but what kind of products will be needed to meet the demands of 1975.

Philip D. Reed, Chairman of the Board (Retired) of General Electric, points out that one estimate indicates that U. S. farmers will need twice as much electricity, appliances, and electrical equipment by 1975. He discusses many farm uses of electricity and quantity of product per man. He may be a little low on some of his figures on expected output per man. His estimate of total broiler production for 1975 was exceeded in 1958. His expected 1½ billion broilers is more likely to be 3 billion or more.

Charles G. Mortimer, Chief Executive of General Foods, indicates some of the probable differences in the diet of tomorrow resulting from better incomes, diet consciousness, and differences in age distribution of the population. He foresees no major problems of meeting tomorrow's food needs. His figure of 25% of disposable income presently spent for food is about 3% higher than those reported by the U. S. Department of Agriculture for recent years.

Charles B. Shuman, President of the Farm Bureau, believes a free market price system will permit production to meet future needs. He does recognize the need for contractual relationships between producers and market operators, however. He also sees the need for controlling inflation and deflation. Non-economists should not be bothered by the statement: " . . . the value of money is impaired by declines in its purchasing power." This is obviously an unintentional redundancy.

Clifford M. Hardin, Chancellor of the University of Nebraska, discusses the role of education in preparing for the future. Among his hopes for the future is recognition of the college community as, not a factory but a unique place of free inquiry, teaching, and seeking for new knowledge.

Herrell F. DeGraff, Food Economist of Cornell University, makes a unique contribution among the group writing for this book by indicating that the real challenge for agriculture is achievement of a level of living for farm families equal to that of families in other economic pursuits.

If one were to be extremely technical, some fault could undoubtedly be found with each of the essays in this book. This does not detract from its worth as a good general survey of the food producing capacity of the country. The book should prove interesting to all students of the food field, and The Ethyl Corporation is to be commended for sponsoring it. Thinking of our hemispheral relations one wishes, however, that in light of the content, "United States" or some other synonym had been used instead of "America" in the title.

R. H. Baker

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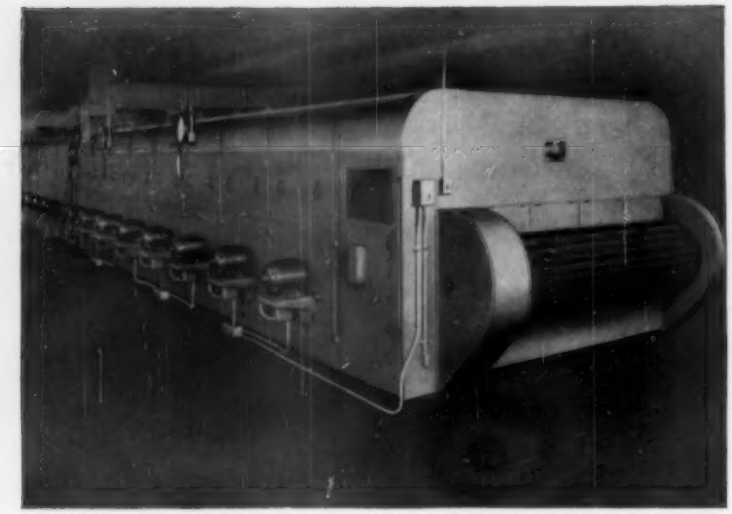

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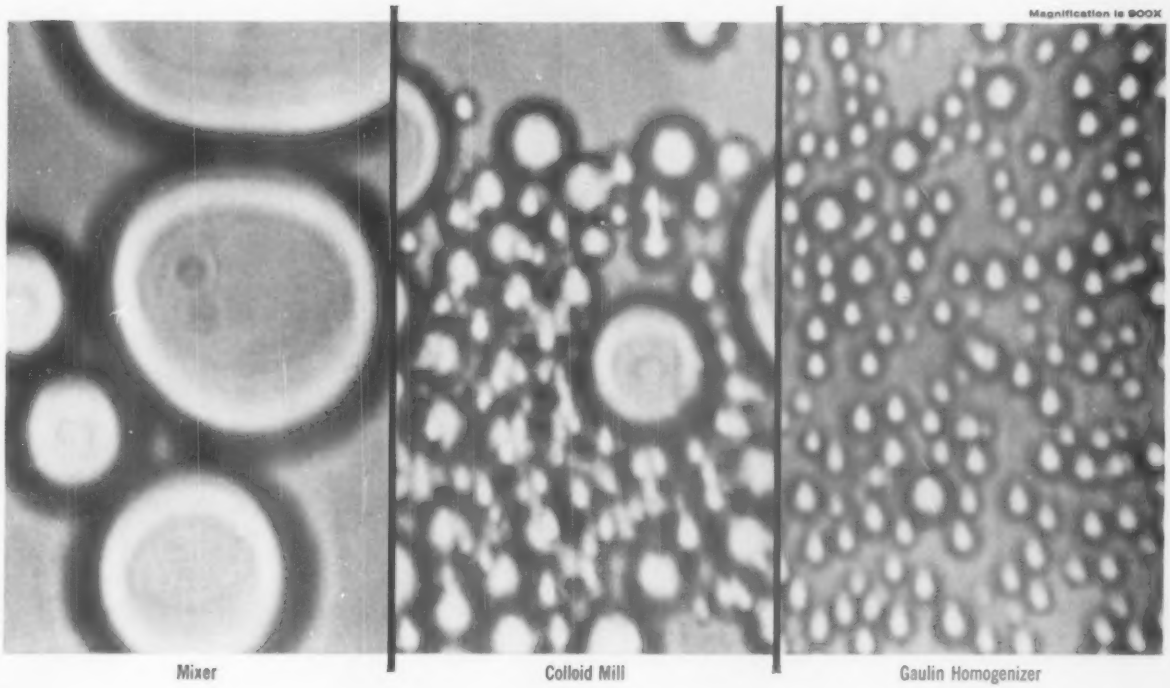

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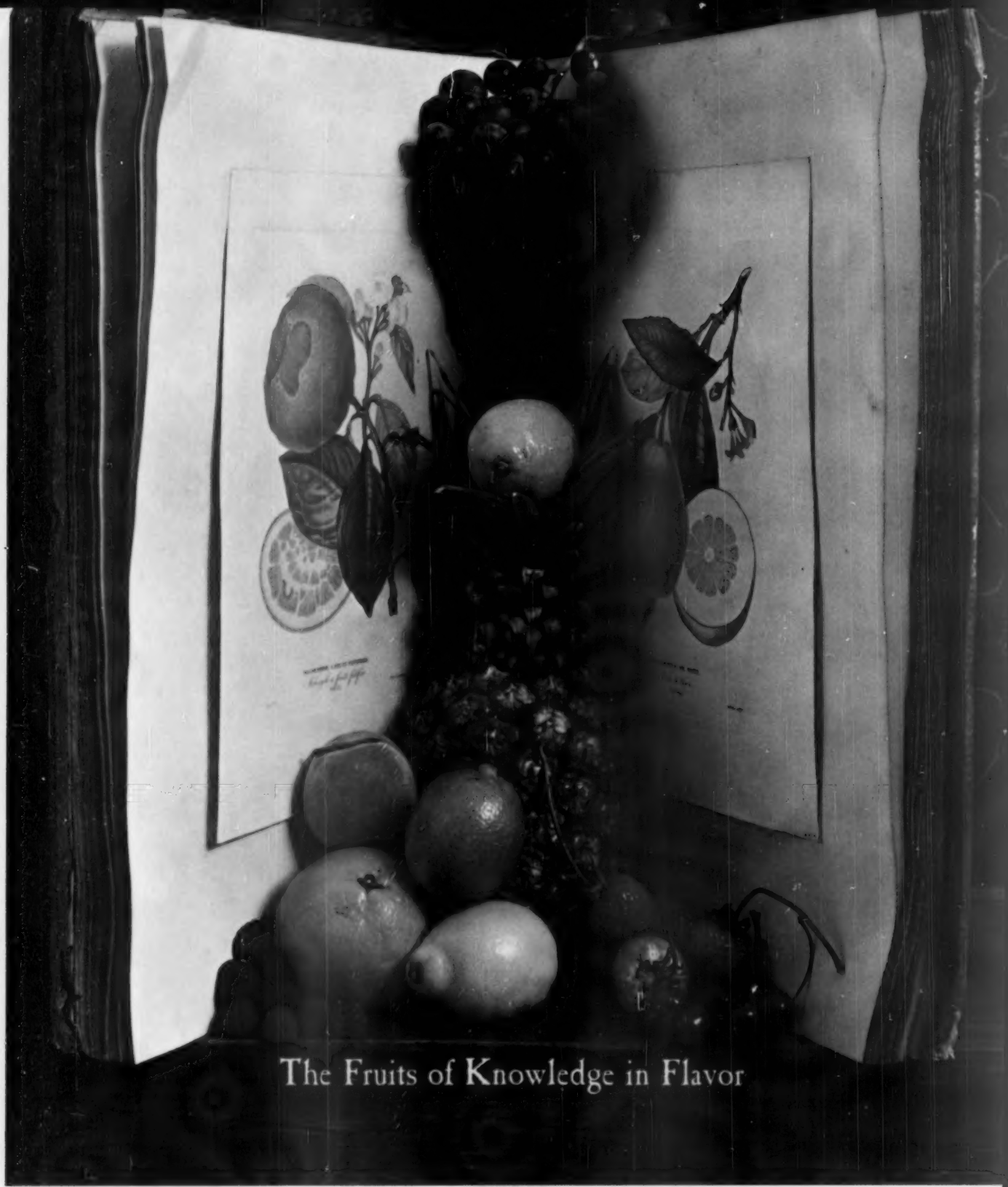

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The Reversibility of the Meat Thawing Process, 1957, O. Shagan, Translated from Russian. Available from the Office of Technical Services, U. S. Department of Commerce, Washington 25, D. C., 50 cents.

BOOKS

Modern Packaging, Encyclopedia issue for 1961, November 1960, Editor, William C. Simms, Packaging Catalog Corp., Emmett Street, Bristol, Conn., U. S. and Canada $3.00, other countries $6.00.

Foods Without Fads, 1960, E. W. McHenry, J. B. Lippincott Company, Philadelphia, New York, $3.50.

PERSONNEL

Continental Can Company, Chicago, has announced the appointment of Rees B. Davis as Senior Research Scientist in Packaging Research, Metal Division Research and Development at Chicago. Dr. Davis has been with the company since 1955 in package development for foods and beverages, and related technical services. He received a Ph.D. degree in Food Technology at The Ohio State University.

R. B. Davis

News Notes

A series of weekly seminars sponsored by the University of California's Department of Food Science and Technology was inaugurated March 17 by David E. Green, University of Wisconsin, speaking on "Mitochondrion and Biochemical Machines."

Dr. Green will be followed each week through May 26 by research scientists from throughout the U. S., Canada and Hawaii.

The series, titled "New Horizons in Food Science," will bring these speakers and subjects to the Davis campus: A. J. Haagen-Smit, California Institute of Technology, "Chemical Components and Food Odors"; W. O. Lundberg, Hormel Institute, University of Minnesota, "Nutritional Studies on Food Lipids"; N. T. Gridgeman, National Research Council (Canada), "Biometry of Sensory Appraisal"; Z. I. Kertesz, New York State Agric. Exp. Sta., Cornell University, "Diet Patterns, Economic Development and Food Technology"; S. N. Timasheff, Eastern Regional Laboratory, USDA, "Physical Chemical Studies of the Associations of the Genetic Species of *beta*-Lactoglobulin"; Samuel Lepkovsky, University of California, Berkeley, "Sensory Properties of Food—What Good Are They?"; Nello Pace, University of California, "Physiological Effects of Alcohol"; Max Levine, Hawaiian Department of Health, "Facts and Fancies of Bacterial Indices in Standards for Waters and Foods"; and B. S. Schweigert, Michigan State University, "Biochemical Changes in Meat During Storage."

The United States Department of Agriculture announced recently, the awarding of a contract to process CCC-owned oats into 5.2 million pounds of rolled oats for distribution to needy families.

The Quaker Oats Co., Cedar Rapids, Iowa, received the contract.

Further strengthening of the curriculum in the Department of Food Science and Technology at the University of California campus comes with the announcement by George F. Stewart, department chairman, of the appointment to the teaching and research staff of Chemical Engineer **Joseph M. Smith,** presently at Northwestern University. Dr. Smith will join the department next summer as Professor of Food Science and Technology, and Chemical Engineer in the Experiment Station.

Dr. Smith, well known for his work in heat transfer and fluid mechanics, will teach a course in engineering principles for food processing operations to upper-division and graduate students, and a graduate course in heat transfer and fluid dynamics. At the same time, he will develop an independent research program on basic and applied engineering science related to food processing, with emphasis on heat transfer and fluid mechanics problems.

The U. C. Department of Food Science and Technology is presently staffed by chemists, biochemists, microbiologists, and technologists dealing with basic and applied investigations into food processing and marketing. Their activities include undergraduate and graduate teaching, and research. The teaching program is heavily oriented toward the graduate level; the research toward basic and applied work on the chemistry, biochemistry, biophysics, and microbiology of foodstuffs and food components. The appointment of Dr. Smith will now expand the research investigations into basic and applied engineering fields.

Dr. Smith is presently professor of chemical engineering, and chairman of the department, at Northwestern University. He previously served as dean of the College of Technology, University of New Hampshire, and on the faculties of the University of Maryland and Purdue. His commercial experience has included engineering posts with The Texas Co., Standard Oil Co. of California, and the Publicker Commercial Alcohol Co., Philadelphia.

Graduating in 1933 from Polytechnic High School, Long Beach, Calif., Dr. Smith received the B.S. degree in Applied Chemistry from the California Institute of Technology in 1937, and the Sc.D. degree in Chemical Engineering from the Massachusetts Institute of Technology in 1943.

Active in several professional societies, Dr. Smith is a member of the American Chemical Society, the American Institute of Chemical Engineers, and the American Society for Engineering Education.

Born in Sterling, Colorado, in 1916, Dr. Smith is the son of Mr. and Mrs. Harold R. Smith, 1006 W. 19th St., Santa Ana, Calif. Smith, his wife, Essie, and their two children will live in Davis.

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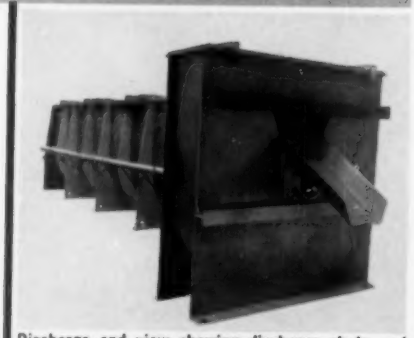

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Chemistry graduate desires position in industry research. He has had extensive experience in meats, but would consider a position as chemist working with other products. Salary, $8,000. REPLY BOX A-2108.

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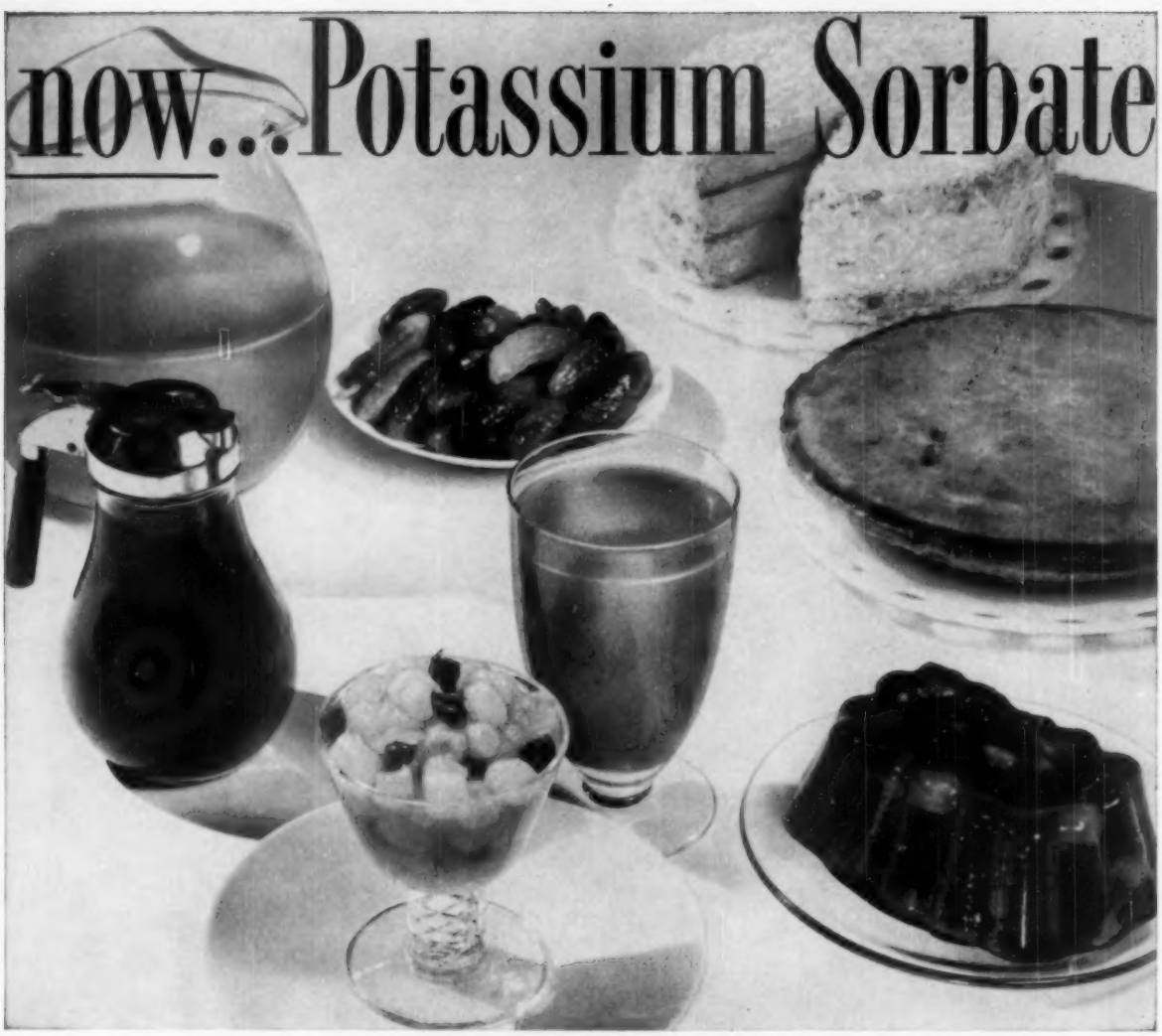

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M.S. with experience in dairy, cereals, and beverages desires position similar to director of development and research. Salary, $15–20,000. REPLY BOX B-4101.

Administrative position desired in area of product development, industry research or quality control by experienced M.S. Salary, $10,800. REPLY BOX B-4102.

Research food technologist position desired by individual with recent M.S. food technology. Product preference includes meats, vegetables and fruits. Salary, $8,000. REPLY BOX B-1111.

M.S. with experience in food manufacturing desires position as food technologist, bacteriologist, or in production. Salary, $8,500. REPLY BOX B-4103.

Technical service position desired by individual interested in dairy products. Interest range also includes quality control and research. Salary, $8,000. REPLY BOX B-5103.

M.S. in food science desires position as production manager. All experience has been in this area. Salary, $11,000. REPLY BOX B-1112.

Another new food technology M.S. with a B.S. in bacteriology desires academic research and/or teaching position, with emphasis on microbiology. She has had two years experience in academic research. Salary, $6,000. REPLY BOX B-1113.

Recent M.S. in microbiology desires industry research, production, or product development position. Primary area of experience is with algae; however fruits, vegetables, fats, oils, and carbonated, distilled and fermented beverages are also encompassed. Salary, $7,000. REPLY BOX B-4401.

Position as Q.C. Manager for Institutional Distributors group desired by B.S. with heavy experience in Q.C. of fruits and vegetables. Salary, $10,000. REPLY BOX A-0101.

Mature individual with heavy experience in product development, production, engineering, and quality control desires production and laboratory supervision position. He holds an Sc.D. in colloid chemistry. Salary, $9,000. REPLY BOX C-2102.

Heavy experience in product development, quality control, and technical service are characteristics set forth by Ph.D. in chemistry who desires to work with flavors, cereals and baked goods on a contract or consulting basis. REPLY BOX A-638

Plant Engineer in processing plant is type of position desired by man with degree in Dairy Industry/Food Technology and Mechanical Engineering. Areas of experience include dairy, and freezing and cooling fresh produce and poultry. Salary, $11,000. REPLY BOX A-0109.

B.S. in Home Economics with major in nutrition and dietetics desires position as home economist with preference towards the area of organoleptic evaluation. She is experienced in meats, soup bases and seasonings. Salary, $7,200. REPLY BOX A-0103.

1950 graduate with B.S. in food technology desires position in product development, production, technical sales, technical service, packaging, or quality control. Salary, $8,000. REPLY BOX A-1115.

Experienced man desires quality control, product development, or production position in the area of vegetables and fruits. Salary, $12,500. REPLY BOX A-5102.

1956 graduate with B.Sc. in Home Economics indicates she is interested in a product development or food promotion position. Salary, $6,200. REPLY BOX A-8401.

Quality control position with primary emphasis on frozen foods desired by 1955 food technology graduate. Salary, $5,800. REPLY BOX A-1116.

Experienced chemistry graduate desires position working with dairy products or cookies. Her position preference is product development, industry research, and quality control. Salary, $8,000. REPLY BOX A-2104.

1957 chemistry graduate interested in cereals, baked goods, and fats and oils, desires production, technical service or quality control position. Salary, $7,200. REPLY BOX A-2105.

Bacteriology graduate desires production or quality control position as food technologist to work with canned foods. Salary, $7,000. REPLY BOX A-4102.

Young Ph.D. desires commercial research and development position which deals with poultry, eggs, or meats. Salary, $8,000. REPLY BOX C-0103.

Line or staff position in manufacturing, packaging or engineering desired by experienced factory management graduate. Salary, $10,000. REPLY BOX A-0104.

Young chemist and biologist desires R & D position in food industry. Experience has been from bench formulations through pilot and plant productions. Salary, $7,000. REPLY BOX A-0105.

Research or teaching position desired by individual with Ph.D. Product preferences include fruits, vegetables, coffee, tea, confections, carbonated beverages, and cereals and baked products. Salary, $9,000. REPLY BOX C-6102.

B.S. degree in Chemistry and Mathematics. 8 yrs. exp. in Statistical Quality Control and as Governmental Food Control Chemist. Some experience with meat. Able to handle correspondence and personnel. Desire position in production, technical service or quality control in South or Southwest. Salary $7,000. REPLY BOX 857.

Man with experience in cereals, baked goods, sugars, syrups, and beverages seeks sales or technical sales position. Has also worked in product development and Q.C. Salary, $7,000. REPLY BOX A-0106.

Q.C. or product development position desired by individual with experience in these areas. Has worked with olives, condiments, sugars, syrups, poultry, eggs, flavors, and colors. Salary, $7,800. REPLY BOX A-0107.

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WANTED: Food Technologists for lab and development work on frozen bakery products. Outstanding opportunity for beginner or man with one or two years experience who can demonstrate promise for growth. In reply specify pertinent training, experience, salary requirements, draft status. Address replies to BOX 856.

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Instructor in research. Midwest location. Background in chemistry, bacteriology, or statistics necessary to work in quantity food research program in a College of Home Economics. REPLY BOX X-2208.

Food Technologist. Midwest location. B.S. in food technology or related areas required. Position entails food product development with all commodities. REPLY BOX X-1312.

Food Technologist/Food Chemist. Eastern location. Several positions open to individuals with B.S. or better and strong background in chemistry. Experience should be with fruits, vegetables, cereals, fish, dairy products, oils, fats and emulsifiers, or flavors. REPLY BOX X-0203.

Assistant Director of Research. Midwest location. Degree in chemistry or food technology with ten years experience in developing food products, and a history of independent operation desired. Position includes new product development in dairy, ice cream, and food vending fields. REPLY BOX X-0305.

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Food technologist—cereal and baked products. Midwest location. B.S. with some graduate work completed. Product development work on cereal products for armed forces rations. REPLY BOX X-0302.

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